VOLUME 16 NUMBER 1

THE ARCHIVES

OF

INTERNAL MEDICINE

JULY 15, 1915

CHICAGO
AMERICAN MEDICAL ASSOCIATION
PUBLISHERS

ENTERED AS SECOND-CLASS MATTER, JANUARY 23, 1908, AT THE POSTOFFICE AT CHICAGO, ILLINOIS, UNDER ACT OF CONGRESS OF MARCH 3, 1879

CONTENTS OF VOLUME XV

JANUARY, 1915. NUMBER 1

FEBRUARY, 1915. NUMBER 2

MARCH, 1915. NUMBER 3

The Archives of Internal Medicine

VOL. XVI. JULY, 1915 No. 1

DIGITALIS DOSAGE*

CARY EGGLESTON, M.D.
NEW YORK

In the course of this investigation I have been struck by the small amount of accurate knowledge that we possess as to practical therapeutics. My experience has been almost exclusively in the laboratory and perhaps I have expected too high a standard in the clinic, but in this field of cardiac tonics alone I see an endless vista of questions to be solved in the clinic if only accurate observations are available. . . . But we have enough of inaccurate therapeutics already; what is needed is not statistical compilation, but an accurate study of each individual case and a careful and, if you will, an experimental investigation of each feature presented.[1]

There are probably no more valuable therapeutic agents in our materia medica than digitalis and its several allies and, although digitalis has been in wide clinical use since Withering first brought it to medical attention, in 1785, the important question of its dosage is still in a state of considerable confusion. Clinicians have recommended doses which range from 2 minims of the tincture (less than ¼ grain of the leaf) three times daily to 30 minims of the tincture (3 grains of the leaf) three times a day. Although the larger dose in this instance is more than twelve times the smaller, this divergence pales into insignificance when compared with that encountered in the case of strophanthus or strophanthin, which Hatcher and Bailey[2] reported from the literature. They found that the largest daily dose used was some 750 times as great in terms of activity as the smallest single dose advised by recognized authorities.

In addition to the gross differences everywhere encountered in the stated doses of the galenical preparations of any of the several members of this group of drugs, the universal practice of expressing the dose in the terms of a measured amount of one or another galenical

* Submitted for publication March 16, 1915.

* From the Department of Pharmacology of the Cornell University Medical School, New York.

1. Cushny, Arthur R.: The Therapeutics of Digitalis and Its Allies, The Harvey Lectures, 1910-1911, p. 46.

2. Hatcher, Robert A., and Bailey, Harold C.: Tincture of Strophanthus and Strophanthin, Jour. Am. Med. Assn., 1909, liii, 5.

preparation fails to take cognizance of the differences in activity of the preparations themselves. That these differences alone are of no small matter is obvious from the reports of many who have tested these preparations. Edmunds and Hale,[3] for example, report variations in the activity of digitalis amounting to 400 per cent., and we have several samples of high grade digitalis leaf in the laboratory, some of which are more than twice as active as others.

The questions of the absorption and excretion, or elimination of the several members of this group and of their preparations have been given little consideration in so far as they enter into the determination of the doses to be used and the frequency of their repetition. The matter of absorption alone is of the utmost importance when the drug is administered orally, and the neglect of this factor has led to much misconception even in the statements of careful and competent investigators. Thus, Cushny[4] and Turnbull[5] both call attention to the fact that the tincture of strophanthus, given orally to man, must be used in larger doses than are given of the tincture of digitalis. Cushny says, "The tinctures of strophanthus and squills were weaker in their action on patients than that of digitalis, nearly twice as much being required to elicit changes in the heart. . . ." And Turnbull, "Although the tincture of strophanthus employed was twenty times as powerful as the digitalis when tested on the frog's heart the dose necessary to produce effects was usually larger."

These statements give the impression that so far as the effect on the human heart is concerned strophanthus is actually less active than digitalis, when each is given by the mouth. That such is not invariably the case, however, is apparent from the fact that in some instances much less is required of the tincture of strophanthus than of digitalis to produce similar effects. That the true explanation of this apparent difference in the activity of the tinctures of strophanthus and of digitalis is referable to the great variability of the absorption of the former from the alimentary canal has been shown for man by Bailey[6] and Hatcher and Bailey,[7] and for the cat by Hatcher.[8]

---

3. Edmunds, Charles W., and Hale, Worth: The Physiological Standardization of Digitalis, Bull. Hyg. Lab., 1908, No. 48.

4. Cushny, Arthur R.: Discussion on the Treatment of Non-Valvular Cardiac Disease, Brit. Med. Jour., 1912, ii, 685.

5. Turnbull, H. H.: The Therapeutic Use of Digitalis, Brit. Med. Jour., 1910, ii, 1608.

6. Bailey, Harold C.: A Clinical Study of Crystalline Strophanthin, Jour. Pharm. and Exper. Therap., 1909-1910, i, 349.

7. Hatcher, Robert A., and Bailey, Harold C.: The Clinical Use of Strophanthus, Jour. Am. Med. Assn., 1910, lv, 1697.

8. Hatcher, Robert A.: Variation of Dosage Dependent on the Method of Administration, Jour. Am. Med. Assn., 1910, lv, 746.

Hatcher and Bailey[7] reported the case of a patient who took 60 minims of tincture of strophanthus (112 cat units) in two days without showing evidence of cardiac action, and who then suddenly developed very threatening toxic symptoms after the seventh dose of 10 minims, or a total of 130 cat units. Hatcher showed that as much as six times the fatal vein dose of ouabain given orally to cats often failed to kill, and when he subsequently tested the animals was able to prove that not even the smallest quantities of the drug had been absorbed in some instances. On the other hand, when as little as twice the minimal fatal vein dose was given orally to a cat a full fatal dose was absorbed in slightly over two and a half hours in one instance.

To complicate the question of the absorption of the digitalis bodies from the alimentary canal of man—and hence to complicate that of dose—we have often to deal with cases of cardiac failure in which the dilatation of the heart has resulted in splanchnic congestion and vomiting when the patients come under treatment.

It is desirable to know whether the diseased heart responds to digitalis quantitatively the same as the normal organ, and whether or not one type of cardiac disease or disorder responds to a smaller dose than another. The influence of age and sex, if any, should be determined, although they are probably of less importance than the several other factors already mentioned.

The last and an important problem is to determine whether or not there is any method of assaying or standardizing the commonly used preparations of the digitalis group of drugs which will yield results that may be transferable to man.

With this brief glance at the more important problems demanding solution before we may be in a position to say that we really know anything about the question of the dosage of digitalis, other than in a general way I will proceed to the presentation of the details of a series of observations bearing on some of these problems which I have been pursuing for the past two years.

PLAN AND SCOPE OF OBSERVATIONS

Inasmuch as the usual method of administering digitalis is by mouth it was thought best to restrict the observations to this mode of giving the drug. The general problem for investigation was to determine whether or not it was possible to establish the dose of digitalis for man on the basis of its activity. With this there were associated several other problems directly concerned in the question of the dosage of this drug and its allies and pure principles. These may be recapitulated as follows:

1. The rate, degree and uniformity of the absorption of the crude drug and its active principles.

2. The influence of sex, age, and weight on the dose.

3. The influence of the preparation—infusion, tincture, etc.—on the dose.

4. The influence of the cardiac condition.

5. The influence of the size of the daily dose on the total dose required.

For the investigation the following preparations were used: (*a*) tinctures made in the laboratory from digitalis leaves from different sources and varying in activity; (*b*) infusions from different leaves and of different activities; (*c*) crystalline digitoxin in solution in 70 per cent. alcohol, or made into tablet triturates with milk sugar; (*d*) true digitalin of Kiliani, and digitalein.

All of these preparations were standardized biologically by the cat method of Hatcher[9] and the activity of each was determined in terms of the cat unit.[10]

The most active tincture used was 2.3 times as potent as the weakest as shown by this method of standardization. The most active infusion was 2.16 times as strong as the weakest, although the infusions were not always prepared from the same leaves as the tinctures.

The same specimen of crystalline digitoxin was used throughout the observations, its activity, therefore, being uniform. However, both the solutions and the tablet triturates were tested after being made, to ascertain their activity.

When the infusions were used they were prepared fresh on the day of the beginning of treatment in each case, and standardized at once. During the time any of the infusions was being given, which never exceeded two days, the preparation was kept tightly stoppered and in the ice box to prevent possible deterioration. Recent experiments have shown us that this precaution was probably unnecessary, as with ordinary care of preparation the infusion should keep for several days at room temperature without material loss of activity.

Owing to certain inherent difficulties in their use the tests of both true digitalin and digitalein were early abandoned, but mention of the former will be made subsequently. The tincture of strophanthus was not used in these observations because of the great variability of

---

9. Hatcher, Robert A., and Brody, J. G.: The Biological Standardization of Drugs, Am. Jour. Pharm., 1910, lxxxii, 360.

10. The cat unit may be defined as that amount of the drug which is just sufficient to kill one kilogram of cat when slowly and continuously injected into the vein. This is expressed in terms of milligrams of the drug, whether it be a pure principle or the leaf.

its absorption from the alimentary canal of man. This fact has already been discussed, and I might here add that, on account of this variability and of its great activity when it is absorbed, I regard its oral administration to man to be so dangerous as to make its use in this way wholly unwarrantable. The same can be said with equal truth of ouabain (crystalline strophanthin of Thoms), the official amorphous strophanthin, and of convallaria. The several other members of the digitalis group of drugs were omitted from consideration because they possess no advantages over digitalis itself, or because they are too infrequently used clinically to warrant their inclusion.

The cases on which the studies were made were unselected so far as their cardiac or general conditions were concerned and included both those in which there was auricular fibrillation and those in which the normal pacemaker was in control of the heart. A few cases were also included in which the heart was not primarily at fault. Two considerations did act to cause some selection, however. The first was that the patient had to be one to whom the drug could be given orally and, with a few exceptions, this excluded patients who were in extremis on admission and who required the immediate injection of ouabain or strophanthin. The second, and the more important consideration for this study, was that it be definitely ascertained that the patient had not received any one of the digitalis group of drugs within a period of not less than three weeks prior to the beginning of observation. It is scarcely necessary to state that no exception was made to this rule.

In every case in which it was possible, without jeopardizing his welfare, the patient was kept in bed without medication other than a possible cathartic or mild hypnotic for a period of from three to seven days before the digitalis or digitoxin was given. During this interval he was weighed at least once, and was given a diet which could be continued without material alteration throughout the course of active treatment. In edematous cases the daily intake of fluids was restricted to a measured amount—usually 40 ounces—and the total amount of urine voided in each twenty-four hours was accurately measured and recorded. In some cases the systolic and diastolic blood pressures were taken daily at the same hour and by the same observer. Such other observations as were demanded in any given case were made and recorded in order to make the conclusions as to the progress of the case under the influence of the drug the more accurate.

During the preliminary period of observation, throughout the time of the administration of the drug, and for a variable period of time after its discontinuance, frequent clinical examinations were made and their results at once committed to writing to reduce error to a mini-

mum. In addition to the information to be gained by careful clinical observation, frequent polygraphic tracings were taken and in some cases electrocardiographic records were also secured.

It was hoped by taking all of these precautions that the element of my personal judgment as to the effects of treatment and the times of their inception might be reduced to a minimum. It must be understood, however, that in the estimation of the progress of any case of cardiac disease the personal element of the observer is bound to play a large and important part, for it is often necessary to interpret

TABLE 1.—CASES SHOWING—

No.	Weight in Lbs.	Age	Preparation No.	Earliest Evidences of Action	Dose in c.c.	Dose in Cat Unit per Lb.
1	193	60	Tr. Dig. 65*.....	Diuresis; clin. impr.†.....	15	0.119
2	160	40	Tr. Dig. 65.....	.....	....	.....
3	150	42	Tr. Dig. 65.....	Clin. impr.	14.65	0.150
4	156	62	Tr. Dig. 90.....	Clin. impr.	12	0.085
5	140	45	Tr. Dig. 105.....	.....	....	.....
6	136	69	Tr. Dig. 148.....	Slight clin. impr.	10	0.049
7	138	59	Tr. Dig. 150.....	Slight clin. impr.	8	0.039
8	127	49	Tr. Dig. 150.....	Marked clin. impr.	19	0.100
9	103	48	Tr. Dig. 150.....	Marked clin. impr.	15	0.097
10	107	58	Inf. Dig. 163.....	.....	....	.....
11	130	55	Inf. Dig. 180.....	Clin. impr.	150	0.096
12	110	68	Inf. Dig. 120.....	.....	....	.....
13	148	43	Digitoxin‡.....	Marked clin. impr.	Mg. 1.5	0.029
14	112	68	Digitoxin.....	Marked clin. impr.	2.5	0.0637
15	117	75	Digitoxin (tab)...	Marked clin. impr.	2.5	0.061

* The number of the tincture or infusion of digitalis is the same as the number of milligrams of
† The abbreviation clin. impr. denotes clinical improvement as defined in the text. H-B. indicates
‡ In the case of digitoxin the cat unit is constant and is 0.35 mg. per kilogram.

any changes observed in the light of previous conditions or of extraneous modifying factors. The independent opinions of the members of the house and visiting staffs of the hospitals in which the work was conducted were solicited, and in a number of cases the decision as to the effect of treatment was reached after a discussion in which several of us took part.

ILLUSTRATIVE CASES

In order to make the method of the study of the cases clearer, abstracts of the essential features in the records of four cases illus-

trating somewhat different conditions and results of treatment are presented.

Patient J. C.—(Case 14 in Table 1), a man aged 68, was admitted to the hospital at about 3 p. m., Oct. 6, 1914, with the diagnosis of chronic cardiac valvular and myocardial diseases and auricular fibrillation. There was no history of antecedent rheumatism. When admitted he was complaining of precordial pain, swelling of his feet and legs, and shortness of breath. His condition was regarded as serious and urgent. The pulse was scarcely palpable at the wrist, very rapid, and extremely irregular in both force and rhythm. Cyanosis was marked and dyspnea and orthopnea extreme. Polygraphic tracings were unsatisfactory, but confirmed the presence of auricular fibrillation.

—Auricular Fibrillation

Full Therapeutic or Minor Toxic Action	Dose in c.c.	Cat Units per Lb.	Total Dose in c.c.	Cat Units per Lb.	Duration of Treatment	Remarks
Nausea; H-B.	25	0.198	30	0.238	4½ days	No emesis from total.
Nausea; C. R.; H-B.	13.3	0.127	.....	.....	16 hrs.	No emesis from total.
Vom.; H-B.	16	0.164	17.5	0.179	2⅔ days	
H-B.	16	0.113	31	0.220	4 days	No emesis from total.
H-B.	16	0.108	.....	.....	36 hrs.	No nausea.
H-B.	26	0.128	32	0.184	4 days	No nausea.
H-B.	30	0.144	.....	.....	4½ days	No nausea.
H-B.	24	0.125	.....	.....	1 day	No nausea.
Nausea; H-B.	35	0.226	.....	.....	4½ days	
H-B.	120	0.103	135	0.115	22 hrs.	Vom. from total.
H-B.	160	0.102	.....	.....	1 day	No nausea.
H-B.	200	0.227	240	0.272	1½ days	No nausea.
H-B.	Mg. 2.5	0.048	.....	.....	3 days	No nausea.
Vom.; H-B.	3.0	0.076	.....	.....	18 hrs.	
Vom.; H-B.	3.5	0.085	.....	.....	1 day	

the leaf in each cat unit.
heart-block, which was partial in all cases. C. R. stands for coupled rhythm.

It was necessary to begin treatment promptly, and Table 2 gives the chronologic course of events in treatment and progress.

At 10 a. m., Oct. 7, after he had received a total of 2.5 mg. of digitoxin in the twelve-hour period from 6 p. m. the evening before to the corresponding hour on the morning of the seventh, his dyspnea was less marked, orthopnea decidedly lessened, and the pulse slower, fuller, and of much better force. The cyanosis had been greatly reduced and the general condition of the patient was very decidedly improved over that of the evening before. On the morning of the eighth scarcely any dyspnea, orthopnea or cyanosis remained; the patient could lie almost flat without respiratory distress, and it was obvious that the full therapeutic effect of the digitoxin had been secured. Tracings taken shortly before the appearance of vomiting showed marked slowing, which was due to the production of a partial heart-block.

Patient P. A.—(Case 5 in Table 2), a man aged 26, was admitted to the hospital June 8, 1914, with the diagnosis of cardiac valvular disease, mitral insufficiency and aortic stenosis. There was a history of previous attacks of rheumatism; the Wassermann reaction was negative. When admitted he was complaining of precordial pain, cough and shortness of breath. Rest in bed with a restricted diet for a week from the time of admission (to June 15) produced little or no change in his condition, and failed to slow his heart. Polygraphic tracings taken at 2 p. m. on the fifteenth showed the rate to be about 92 per minute with the rhythm normal. At midnight, June 15, he was given 8.0 c.c. of tincture of digitalis No. 97 at a single dose. Ten hours later (10 a. m. on the sixteenth) tracings showed the pulse rate to be only 81 per minute, a drop of 11 beats, and the rhythm was then distinctly coupled. The patient said that he was much more comfortable. During the day of the sixteenth the precordial pain and dyspnea almost disappeared. The cough rapidly subsided and although no further digitalis was given, the patient made a rapid recovery and was discharged June 27.

TABLE 2.—Chronologic Course of Events in Treatment of J. C.

Date 1914	Hour	Digitoxin, Mg.	Heart Rate	Radial Pulse	Deficit	Total Urine in 24 Hrs.	Remarks
10/ 6	4 p.m.	.....	170	135	35	.....	
	6 p.m.	1.5					
	12 p.m.	0.5					
10/ 7	6 a.m.	0.5	.....	.....	....	18 ounces	14 hours only.
	10 a.m.	.....	94	88	6	.....	Marked improvement.
	12 m.	0.5					
	12:45 p.m.	.....	.....	.....	....	.....	Vomited; heart-block.
	3:30 p.m.	.....	91	83	8		
10/ 8	10 a.m.	....	94	85	9	53 ounces	
	4:30 p.m.	.....	92	88	4		
10/ 9	9 a.m.	.....	86	82	4	41 ounces	
10/10	9:30 a.m.	.....	82	75	7	73 ounces	

Patient B. B.—(Case 33 in Table 3), a woman aged 49, was admitted to the hospital Oct. 20, 1914, with the diagnosis of chronic cardiac myocardial and valvular diseases; mitral insufficiency; hepatic cirrhosis. When admitted she was complaining of swelling of abdomen, shortness of breath, and cardiac palpitation. Her pulse was rapid and of poor force but regular in rhythm; there was considerable edema and ascites, and the respiration was rapid and embarrassed. Free catharsis, diet with fluid intake restricted to 40 ounces in the twenty-four hours, and rest in bed were without beneficial effect, although continued for five days. Tablet triturates, each containing 0.5 mg. of digitoxin, were given from November 4 to 6 as follows: November 4 at 6 p. m. two tablets (1.0 mg.); one tablet at midnight (0.5 mg.); November 5 one tablet every six hours to and including the 6 a. m. dose on November 6. The total taken was 4.0 mg. in a period of thirty-six hours.

A note made at 11:30 a. m., November 5, states: "No demonstrable evidence of digitoxin action after 2.0 mg. in twelve hours." And a note made at 10 the following morning states: "Very slight sinus arrhythmia this morning. No other signs of digitalis action on the heart except stronger and fuller pulse. Has had total of 4.0 mg. of digitoxin. Complains of slight nausea which was

first noticed yesterday afternoon after a total of 2.5 mg. digitoxin had been taken." Although the patient vomited twice on the afternoon of November 5 there was still no influence of the digitoxin on the rate of her heart and the drug produced no beneficial action.

Patient F. C.—(Case 34 in Table 3), a man aged 37, was admitted to the hospital Nov. 13, 1914, complaining of swelling of scrotum and legs, weakness on exertion, dyspnea, orthopnea, and cardiac palpitation. Diagnosis: Chronic parenchymatous nephritis, pericarditis with effusion, cardiac hypertrophy and dilatation, mitral insufficiency.

Tablet triturates, each containing 0.5 mg. of digitoxin, were given as follows: November 17, two at 6 p. m. and again at midnight; November 18 at 6 a. m., one tablet, and this dose was then ordered to be repeated every six hours night and day and its administration thus continued until the 6 a. m. dose was given on November 20, making a total of 6.5 mg. in sixty hours.

The following indications of the progress of this case are taken from notes made at the time:

November 17, 9:30 a. m.: No improvement in the patient's condition has resulted from rest and the restriction of the intake of fluid to not over 40 ounces in twenty-four hours. The edema is still very marked, the pulse continues rapid, and dyspnea and orthopnea still trouble him. Polygraphic tracings show regular rhythm, disturbed only by a slight grade of sinus arrhythmia which is for the most part synchronous with respiration.

November 18, 10 a. m.: Pulse rate and rhythm continue unchanged since yesterday, but the dyspnea is decidedly less and the urinary output has increased. This after 2.5 mg. of digitoxin in twelve hours.

November 19, 9:30 a. m.: Clinical improvement is very marked, the pulse and respiration rates range somewhat lower, both the dyspnea and orthopnea are much diminished and diuresis is marked. There is no nausea. The patient has now taken 4.5 mg. of digitoxin in thirty-six hours and the full therapeutic effects of the drug are becoming manifest.

November 20, 10 a. m.: Patient nauseated early last night and he has vomited twice. The vomiting came on after 5.5 mg. of digitoxin had been taken, but the administration was continued to the 6 a. m. dose, making a total of 6.5 mg. in sixty hours. Diuresis very marked, edema nearly gone, respiration slower, no orthopnea. Pulse slowed through partial heart-block, the fourth or fifth ventricular beat being regularly blocked. The observations recorded in the course of treatment of this patient are graphically presented in the accompanying chart.[11]

In the presentation here given of these four cases all but the essential features have been omitted for the sake of brevity. All other points were observed which contributed to the formation of a conclusion as to the exact status of the individual patient at the time of each examination.

A total of fifty-three courses of treatment were followed in this study and comprise the material shortly to be subjected to analysis. These fifty-three courses were carried out on forty-seven patients, there being six instances in which one patient either remained in the hospital long enough to require a second or third course, or returned

11. The very large dose of 6.5 mg. of digitoxin given to this patient was administered under exceptional conditions and, though the patient made a satisfactory and complete recovery without untoward incident, such a large dose is distinctly not advised.

TABLE 3.—

No.	Weight in Lbs.	Age	Preparation No.	Earliest Evidences of Action	Dose in c.c.	Dose in Cat Unit per Lb.
1	127	25	Tr. Dig. 65*.....	Clin. impr.; S. A.†.....	12	0.145
2	165	52	Tr. Dig. 90.....	.....	....	.....
3	134	28	Tr. Dig. 97.....	Slight clin. impr.	12	0.091
4	129	60	Tr. Dig. 97.....	Diuresis; clin. impr.	10	0.080
5	121	26	Tr. Dig. 97.....	.....	....	.....
6	158	42	Tr. Dig. 97.....	Diuresis; clin. impr.; S. A.	8	0.052
7	120	63	Tr. Dig. 97.....	Diuresis; clin. impr.; S. A.	8	0.069
8	144	60	Tr. Dig. 97.....	Clin. impr.; S. A.	16	0.114
9	144	60	Tr. Dig. 97.....	Clin. impr.; S. A.	16	0.114
10	148	34	Tr. Dig. 97.....	Clin. impr.	6	0.041
11	141	49	Tr. Dig. 97.....	Marked clin. impr.	16	0.116
12	132	64	Tr. Dig. 97.....	Clin. impr.; S. A.; ex. syst.	12	0.092
13	172	56	Tr. Dig. 97.....	Clin. impr.; S. A.; diuresis.....	17	0.101
14	129	54	Tr. Dig. 105.....	.....	....	.....
15	143	54	Tr. Dig. 130.....	Marked clin. impr.	17	0.091
16	140	48	Tr. Dig. 148.....	Clin. impr.	25	0.120
17	100	48	Tr. Dig. 150.....	Clin. impr.; slowing.....	20	0.133
18	52	11	Inf. Dig. 83.....	.....	....	.....
19	131	61	Inf. Dig. 110.....	.....	....	.....
20	145	60	Inf. Dig. 110.....	Clin. impr.; S. A.	100	0.093
21	52	8	Inf. Dig. 120.....	.....	....	.....
22	145	32	Inf. Dig. 180.....	Clin. impr.; diuresis; S. A.	125 Mg.	0.072
23	133	45	Digitoxin‡.....	Slight clin. impr.; S. A.	1.5	0.032
24	123	45	Digitoxin.....	Clin. impr.; slowing.....	2.0	0.045
25	131	60	Digitoxin.....	.....	....	.....
26	131	60	Digitoxin.....	Clin. impr.; slowing.....	1.25	0.027
27	165	59	Digitoxin.....	Clin. impr.; diuresis.....	1.5	0.026
28	154	40	Digitoxin.....	No clin. impr.; ex. syst.	3.0	0.055
29	113	39	Digitoxin.....	Clin. impr.; S. A.	1.25	0.0315
30	113	39	Digitoxin.....	Clin. impr.; S. A.	1.25	0.0315
31	151	46	Digitoxin.....	Clin. impr.; S. A.; H-B.	2.5	0.047
32	145	52	Digitoxin.....	No clin. impr.; S. A.	1.75	0.034
33	130	49	Digitoxin (tab)...	.....	....	.....
34	162	37	Digitoxin (tab)...	Clin. impr.; diuresis.....	2.5	0.044
35	145	60	True digitalin‖....	Clin. impr.; slowing.....	8.0	0.036
36	142	34	True digitalin.....	Slight S. A. only.....	16.0	0.075
37	93	38	Digitalein¶.....	.....	....	.....
38	93	38	Digitalein.....	Slowing only	48.0	0.147

* In this table the number of the digitalis is the same as the number of milligrams in one cat unit.
† Clin. impr. denotes clinical improvement as defined in text. H-B. indicates heart-block, which was indicates clinical evidence of full effect.
‡ The cat unit of digitoxin is 0.35 mg. per kilogram.
‖ The cat unit of true digitalin is 1.5 mg. per kilogram.
¶ The cat unit of digitalein is 3.5 mg. per kilogram.

—Nonfibrillating Cases

Full Therapeutic or Minor Toxic Action	Dose in c.c.	Cat Unit per Lb.	Total Dose in c.c.	Cat Unit per Lb.	Duration of Treatment	Remarks
Clin. ev.; S. A.	14	0.169	.....	.....	1 day	No nausea.
Vom.; slight clin. impr. ...	47	0.293	59	0.368	5 days	
Vom.; slight clin. impr. ...	24	0.182	.....	.....	1¾ days	Pulmonary tuberculosis.
Clin. ev.; diuresis.....	16	0.127	44	0.350	6 days	Vom. from total.
Clin. ev.; slowing; C. R. ...	8	0.068	.....	.....	.....	Single dose.
Diuresis; slowing; S. A. ...	24	0.150	.....	.....	3 days	First effect after single dose.
Diuresis; slowing	10	0.086	.....	.....	18 hrs.	First effect after single dose.
Clin. ev.; slowing; S. A. ...	32	0.228	.....	.....	4 days	No nausea.
Clin. ev.; slowing; S. A. ...	18	0.128	.....	.....	3 days	Same case as No. 8.
Clin. ev.; slowing.....	12	0.082	.....	.....	30 hrs.	First effect after single dose.
Nausea; clin. ev.	26	0.190	28	0.200	3½ days	No vom.
H-B.; clin. ev.	18	0.138	26	0.200	60 hrs.	No vom.
S. A.; clin. ev.	29	0.174	.....	.....	2½ days	No nausea.
S. A.; clin. ev.	7	0.051	.....	.....	.....	Single dose.
Nausea; diuresis; S. A.	25	0.133	.....	.....	36 hrs.	
Clin. ev.; H-B.	35	0.168	45	0.217	3 days	Naus. after 39 c.c. = 0.18 c. u. per lb.
Mod. clin. impr.	66	0.440	.....	.....	5 days	No toxic symptoms.
S. A.	30	0.104	.....	.....	28 hrs.	No clin. impr.
Vom.; no clin. impr.	140	0.145	180	0.187	36 hrs.	Vom. only once.
Naus.; clin. ev.; S. A.	200	0.186	.....	.....	42 hrs.	
Naus.; no clin. impr.	55	0.130	.....	.....	12 hrs.	
Clin. ev.; S. A.; slowing....	230 Mg.	0.132	.....	.....	2 days	No nausea.
Clin. ev.; S. A.; slowing....	3.625	0.077	.....	.....	5½ days	No nausea.
Nausea; clin. ev.	4.75	0.110	5.5	0.127	7 days	Vom. after total.
Nausea; H-B.; ex. syst.	1.75	0.038	.....	.....	1½ days	Same case as No. 19.
H-B.; ex. syst.	2.25	0.049	.....	.....	2 days	Same case as No. 19.
Naus.; diuresis, slowing....	3.50	0.060	.....	.....	64 hrs.	No vomiting.
No clin. impr.; ex. syst.	3.00	0.055	4.0	0.073	3½ days	No nausea.
Clin. ev.; S. A.	2.50	0.063	3.5	0.088	2½ days	Naus. after total.
Clin. ev.; S. A.	2.50	0.063	.....	.....	1½ days	No nausea. Same as No. 29.
Vom.; clin. ev.; H-B.	3.50	0.066	.....	.....	1½ days	
Naus.; clin. ev.; S. A.	3.50	0.068	.....	.....	2 days	
Naus.; no clin. impr.	2.50	0.054	4.0	0.087	1½ days	Vom. after total.
Diuresis; S. A.; slowing....	4.50	0.079	6.5	0.114	60 hrs.	Vom. after 5.5 mg. = 0.096 c. u. per lb.
Clin. ev.; S. A.	28.0	0.128	.....	.....	3 days	No nausea. Same as No. 20.
Diuresis; slight S. A.	180.0	0.840	.....	.....	4¼ days	No nausea.
Slowing; ex. syst.	36.0	0.110	.....	.....	44 hrs.	No nausea.
Slowing; ex. syst.	64.0	0.196	.....	.....	3 days	Same case as No. 37.

partial in all cases. S.A. stands for sinus arrhythmia. C. R. stands for coupled rhythm. Clin. ev.

to the hospital within a few months after discharge. For the sake of brevity and to obviate the necessity of the reader's having to search through the abstracts of case histories, the essential features of each case, so far as the present problems are concerned at least, are presented in Tables 1 and 3. In this form the observations lend themselves more readily to analysis, and it is from these tables that most of my later figures will be drawn. In these tables it has been impos-

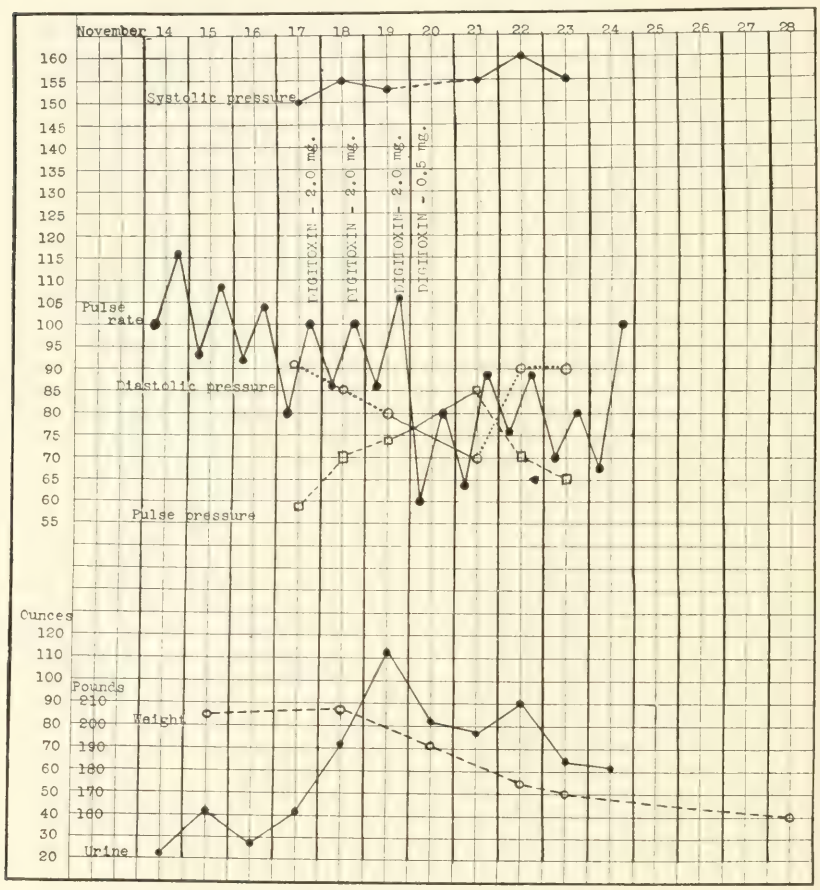


Chart showing course of events in treatment of Patient F. C.

sible to do more than to indicate the type of phenomena present at any of the stages of action recorded, and the significance of some of the terms employed needs definition.

CLINICAL IMPROVEMENT

Exception may be taken to the use of a term apparently so loose as this in a paper in which an effort is to be made to arrive at

numerical results. The term is here employed to express the sum total of all of the factors which go to make up the purely clinical features indicative of improvement, exclusive of all traceable cardiac phenomena. Such clinical features differ in type with the different cases and they include both subjective and objective phenomena. Among the former some of the most trustworthy are the patient's voluntary statements as to improvement in his respiration; the ability to lie flatter in bed without suffering respiratory oppression; disappearance of the sensation of epigastric fulness, distress, or pain; disappearance of nausea or vomiting due to congestion; loss of precordial and referred pains; and the disappearance of cardiac palpitation.

The more usual objective symptoms of value are a fall in the pulse and respiration rates; the more or less rapid subsidence of dyspnea and orthopnea; disappearance of persistent cough, and the clearing up of the signs of congestion at the lung bases. The gradual decrease in the pulse deficit in cases of auricular fibrillation; more or less rapid disappearance of the signs of a congested liver and splanchnic region such as, loss of tenderness over the enlarged liver or the epigastrium, diminution in the size of the liver, disappearance of its pulsation, and the vanishing of tympanites are all signs of first importance. So also are the clearing up of cyanosis and cold extremities, the disappearance of edema, and an increase in the urinary output.

Such symptoms as these, in varying combinations, go to make up the picture denoted "clinical improvement," and in some cases in which one symptom stood out most prominently it has been indicated in the table. So, also, the clinical improvement has been indicated according to degree as slight, moderate, or marked.

The classification which includes all such phenomena as marked sinus arrhythmia, partial heart-block, extra systoles, etc., as minor toxic effects of digitalis, has been adopted in this discussion. In addition to these both nausea and vomiting have been included as minor toxic symptoms because they appear so frequently in close association with one or another of the previously mentioned phenomena. Further all of these strictly *minor* toxic phenomena appear, in cases amenable to benefit by digitalis, so nearly simultaneously with the development of the fullest obtainable therapeutic effects that these two sets of phenomena have been grouped under the same rubric, although there will be occasion to separate them for subsequent analysis.

All of this attention has been devoted to the presentation and elucidation of the plan and scope of these observations in order to enable the reader to follow the ensuing discussion of the results the more easily, and to place him in a position to analyze the findings for himself in any way he may desire. I might say that I arrived at just

this point in the actual conduct of my work on the problems in hand without any very definite idea of what the observations would show, for I made no analysis of them until the work of observation was completed. In this way I hoped to avoid the pitfall of the preconceived idea.

ANALYSIS OF OBSERVATIONS

As previously stated, the material for such analyses as will now be undertaken is set forth in Tables 1 and 3. It will be seen that each table gives the weight of the patient in pounds, the age of the patient, the preparation of digitalis which was used, together with its activity in terms of the cat unit, the actual amount of the preparation required to produce the several types of effects recorded, the amount of each preparation similarly required calculated on the basis of activity in fractions of a cat unit per pound of the body weight of the patient, and finally, the total period of time during which the drug was administered, with certain remarks on individual cases which were deemed of value for their better understanding. With these tables available for reference we may proceed at once to the discussion of certain of the phases of the question in hand.

AVERAGE DOSES WITH MAXIMUM VARIATIONS FROM AVERAGE, IN FRACTIONS OF A CAT UNIT OF ACTIVITY PER POUND OF BODY WEIGHT OF PATIENT

These doses were determined in the following manner: The actual amount of the preparation administered up to the time of production of the effect in question was known, and this was reduced to terms of milligrams of the leaf in the case of the galenical preparations of digitalis. The cat unit of activity for each preparation had been established and, as this is always stated in terms of milligrams of digitalis leaf irrespective of the preparation, the total number of cat units which the patient received could be readily determined by division of the total number of milligrams by the number in each cat unit. The total number of cat units which were taken was then divided by the weight of the patient in pounds and the resulting figure gave the fraction of a cat unit which had been actually taken per pound of body weight to produce the effect in question. With the pure principles the first step was not required, for they were always given in terms of milligrams of the dry substance. The fractions of a cat unit per pound actually taken by each patient are recorded in the tables for each effect produced, and from these figures Tables 4 and 5 have been prepared in which are set forth the average doses for three preparations—tincture and infusion of digitalis, and crystalline digitoxin—for the production of each effect recorded, together with the maximum

and minimum doses which gave similar effects in any case, the range of variation from the average, and the number of cases entering into the computation of the average doses.

Table 4 gives the doses in the fifteen cases of auricular fibrillation. A larger number of observations would have been desirable but more patients fulfilling the strict requirements were unfortunately not available. Even with this relatively small number of cases the results are far more uniform than might reasonably have been expected. It must be borne in mind that there is a great difference between determining the fatal dose of a drug for one of the lower animals by intravenous administration or subcutaneous injection and attempting to determine

TABLE 4.—Average Doses in Fractions of a Cat Unit per Pound of Body Weight, with Maximum and Minimum Doses and the Percentage Variations, in Cases with Auricular Fibrillation

Effect Produced	Preparation	No. of Cases	Average Dose	Maximum Dose	Percentage Above	Minimum Dose	Percentage Below
Earliest.....	Tinctures	7	0.091	0.150	64	0.039	58
	Infusions	1	0.096				
	Digitoxin	3	0.051	0.0637	24	0.0289	44
Therap. without naus. or vomit.	Tinctures	5	0.123	0.144	17	0.108	13
	Infusions	3	0.144	0.227	57	0.102	25
	Digitoxin	1	0.048				
Nausea or vomiting...	Tinctures	4	0.179	0.226	26	0.127	30
	Infusions	1	0.115				
	Digitoxin	2	0.080	0.085	..	0.076	
Therapeutic, all cases	Tinctures	9	0.148	0.226	51	0.108	26
	Infusions	3	0.144	0.227	57	0.102	13
	Digitoxin	3	0.069	0.085	23	0.048	31
Total taken.....	Tinctures	4	0.205	0.238	16	0.179	13
	Infusions	2	0.193	0.272	..	0.115	

a dose for man when the drug is given by mouth, in which case the factors of absorption may play an important rôle in modifying the dose. And, further, it must be remembered that in the case of the human subject we have no sharply fixed and constant end-point which can be reached in all cases, but that on the contrary the end-point for each determination of dose must of necessity be somewhat different in any two cases. The determination of just that dose which may be regarded as effective in any given case, must depend on what, in that case, one is compelled to take as the index of effectiveness. The attempt to determine the several doses on man as compared to the determination of the activity of the same preparations on the lower animals presents a certain analogy to the performance of two titrations, in the one case using an indicator with no very sharp color change to mark the end reaction, in the other using one with a very decided and sharp change of color which appears precisely at the

change in reaction. It is no wonder, therefore, that the figures should show some range of variation. Indeed, it seems remarkable that the variation was not very much greater than has actually been found.

From Table 4 we see that the greatest range of variation from the average dose for both the tincture of digitalis and for digitoxin was encountered with that dose which produced the earliest evidences of action. This was to be anticipated under existing conditions. The first effects produced are not necessarily alike in any two cases, even among fibrillating hearts. But more important is the fact that it was my practice to give relatively large initial doses with the earlier repetitions also of quite large size, so that it is more than likely that in a few cases somewhat smaller amounts might have given evidence of some action. But even here the extreme range of variation from the average in seven cases receiving the tincture of digitalis is only 122 per cent. of the average dose—64 per cent. above in one case, and 58 per cent. below in another—but reference to Table 1 will show that the doses taken by four of the seven patients for the production of earliest effects differed very slightly from the average of 0.091 cat unit per pound.

Where nausea or vomiting was taken as the effect for which the dose was to be determined the variation from the average was found to be much less than in the case of the earliest effects. Thus, of four patients developing these symptoms after taking the tincture of digitalis, one required a dose 26 per cent. above the average and another developed the symptom with a dose 30 per cent. below the average. Only two patients vomited from digitoxin, and the doses producing emesis lay very close together.

Under the caption, "therapeutic, all cases" are included all of the cases recorded in the columns devoted to the full therapeutic or minor toxic actions in Table 1. The average dose of tincture of digitalis required to produce these effects was 0.148 cat unit per pound, and the largest and smallest doses recorded show a variation of 51 per cent. above and 26 per cent. below the average, or a total range of variation of 77 per cent. The doses determined for three patients receiving infusion of digitalis lay very close to those just stated for the tincture, in average, maximum, and minimum. The total range of variation from the average dose was decidedly less with digitoxin, amounting to only 54 per cent. This narrower range is possibly due in part to the small number of cases receiving digitoxin in this series.

Turning to Table 5, which embodies the corresponding observations made in cases without auricular fibrillation, we find both a larger number of observations and a rather wider range of variation, in isolated cases, from the averages determined. From fourteen courses of

administration of the tincture an average dose of 0.097 cat unit per pound was established for the production of the earliest effects, and the total variation from this average rose to 121 per cent.—one case requiring 49 per cent. more than the average, and one responding to 72 per cent. less. As in the previous series of observations, here too, the range of variation from the average in the case of digitoxin is found to be smaller than in the case of the tincture of digitalis, being only 78 per cent. total—48 per cent. above and 30 per cent. below.

For the production of nausea or vomiting the average doses were determined from six patients who received the tincture, three the infusion, and eight digitoxin. The total range of variation in the

TABLE 5.—Average Doses in Fractions of a Cat Unit per Pound of Body Weight, with Maximum and Minimum Doses and the Percentage Variations, in Nonfibrillating Cases

Effect Produced	Preparation	No. of Cases	Average Dose	Maximum Dose	Percentage Above	Minimum Dose	Percentage Below
Earliest.....	Tinctures	14	0.097	0.145	49	0.041	72
	Infusions	2	0.082	0.093	..	0.072	
	Digitoxin	10	0.037	0.055	48	0.026	30
Therap. without naus. or vomit.	Tinctures*	12	0.130	0.223	75	0.051	61
	Infusions	2	0.118	0.132	..	0.104	
	Digitoxin	6	0.064	0.079	23	0.049	24
Nausea or vomiting...	Tinctures	6	0.221	0.350	58	0.133	40
	Infusions	3	0.153	0.186	21	0.130	16
	Digitoxin	8	0.072	0.110	52	0.038	48
Therapeutic, all cases	Tinctures†	16	0.148	0.293	97	0.051	66
	Infusions	5	0.139	0.186	33	0.104	26
	Digitoxin	12	0.065	0.110	69	0.038	42
Total taken.....	Tinctures‡	5	0.267	0.368	37	0.200	26
	Infusions	1	0.187				
	Digitoxin	5	0.096	0.127	32	0.073	23

* Exceptional Case 17 omitted. If included, average becomes 0.154 cat units per pound.
† Exceptional Case 17 omitted. If included, average becomes 0.165 cat units per pound.
‡ Exceptional Case 17 omitted. If included, average becomes 0.295 cat units per pound.

case of the tincture was 98 per cent. of the average dose; in that of the infusion 37 per cent. (only three cases); and in that of digitoxin 100 per cent.

When the production of the full therapeutic or minor toxic actions is considered the total variation in the case of the tincture rises abruptly, on account of a single case, to 163 per cent., but attention is to be called to the fact that this greater variation is not due so much to the occurrence of instances of greater susceptibility to the effects of digitalis as to those of abnormal tolerance.[12] Administration of the

12. I have encountered one instance of very great tolerance to the effects of digitalis in which 0.44 cat unit per pound induced only moderate clinical improvement without any toxic symptoms. Whether or not this was due to defective absorption I am unable to state. I have not included this case in the computation of any of the averages here recorded. The details will be found under No. 17 in Table 3.

infusion to the production of full therapeutic effects in five cases gave results in which the variation was relatively slight, the total being only 59 per cent. of the average dose. The results obtained from administration of digitoxin to twelve patients to the same point fall into an intermediate position between those recorded for the tinctures and infusions. Thus the total variation is here found to be 111 per cent. of the average dose.

Tables 4 and 5 give the complete data along these lines of analysis for all of the cases in each of the two groups, but sufficient illustrations have been drawn from them to indicate the ordinary ranges of

TABLE 6.—Average Doses in Fractions of a Cat Unit per Pound of Body Weight, with Maximum and Minimum Doses and the Percentage Variations, in all Cases Observed, Regardless of Presence or Absence of Fibrillation

Effect Produced	Preparation	No. of Cases	Average Dose	Maximum Dose	Percentage Above	Minimum Dose	Percentage Below
Earliest.....	Tinctures	21	0.095	0.150	57	0.039	59
	Infusions	3	0.087	0.096	10	0.072	18
	Digitoxin	13	0.040	0.0637	59	0.026	35
Therap. without naus. or vomit.	Tinctures*	17	0.128	0.228	78	0.051	60
	Infusions	5	0.133	0.227	70	0.102	24
	Digitoxin	7	0.062	0.079	27	0.048	23
Nausea or vomiting...	Tinctures	10	0.204	0.350	71	0.127	38
	Infusions	4	0.144	0.186	29	0.115	20
	Digitoxin	10	0.074	0.110	48	0.038	49
Therapeutic, all cases	Tinctures†	25	0.148	0.293	97	0.051	66
	Infusions	8	0.141	0.227	69	0.102	28
	Digitoxin	15	0.066	0.110	66	0.038	43
Total taken.....	Tinctures‡	9	0.239	0.368	53	0.179	26
	Infusions	3	0.191	0.272	42	0.115	40
	Digitoxin	5	0.096	0.127	32	0.073	23

* Exceptional Case 17 omitted. If included, average becomes 0.146 cat units per pound.
† Exceptional Case 17 omitted. If included, average becomes 0.159 cat units per pound.
‡ Exceptional Case 17 omitted. If included, average becomes 0.259 cat units per pound.

variation which are encountered in the determination of the human doses of digitalis and digitoxin.

In Table 6 all of the observations made with tinctures and infusions of digitalis and with digitoxin are brought together irrespective of the question of fibrillation. In this table, which is virtually a composite of the preceding two, the greatest variations from the average doses do not differ very materially from those already discussed, although this grouping together of all of the cases somewhat increases the range of variation.

We have so far dealt only in terms of the greatest variations from the averages in the several instances and have thus placed our subsequent remarks regarding the actual dosage of digitalis for man, as based on our figures, in the least favorable light. For detailed con-

sideration we will confine our attention to the dose which was found necessary for the production of the full therapeutic effects or of one or another of the minor toxic actions of the drug, as this is the dose which we usually desire to give to any patient who requires treatment by digitalis.

THE FULL THERAPEUTIC DOSE IN FRACTIONS OF A CAT UNIT OF ACTIVITY PER POUND OF BODY WEIGHT

Reference to the average doses stated in tables 4, 5 and 6 for the tincture and the infusion of digitalis and for digitoxin, respectively, in the group classified as "therapeutic, all cases" (including both those showing full therapeutic and those with minor toxic actions) will at once make it evident that there is little essential difference between the doses of the respective preparations dependent on the presence or absence of auricular fibrillation, and the averages for any preparation for all of the cases combined are essentially the same as those for each of the two divisions taken separately. We will therefore first turn our attention to the averages of all the cases, and take up the two main classes later.

If Table 6 be taken in conjunction with Tables 1 and 3 for details, we can make the following deductions:

1. The average dose of the tincture of digitalis required to produce full therapeutic effects or minor toxic actions is 0.148 cat unit per pound of the patient's body weight. This is based on the results of 25 courses of administration of tinctures of widely different relative activities. In eight of these the dose producing these effects fell below the average to an extent not exceeding 15 per cent. of the average dose. In five others the dose required exceeded the average by an amount not greater than 15 per cent. of the average dose. In other words, the doses taken in 13, or 52 per cent. of the cases were divergent from one another by the extreme limit of 30 per cent. of the average dose. One resistant patient required 97 per cent. more than the average dose for the production of effects, while the most susceptible one responded to a dose 66 per cent. below the average.

2. The average dose of the infusion of digitalis was found to be 0.141 cat unit per pound of the patient's body weight. Eight patients were studied in this determination and they received infusions differing considerably in strength. Of these eight patients, two responded to doses not over 15 per cent. less than the average dose, and one required more than the average by an amount not exceeding 15 per cent. On the other hand, one patient required an excess of 69 per cent. over the average to cause the desired effects, while another was susceptible to the extent of showing the full effects from a dose less than the average by 28 per cent.

3. The average dose of 0.066 cat unit per pound of the patient's body weight was established for crystalline digitoxin as the result of its administration to fifteen patients. Of these there were five in whom the response was secured with a dose below the average by an amount not exceeding 15 per cent. and a dose not more than 15 per cent. above the average was required for the production of effects in three cases. That is, eight cases, or 53 per cent. responded to doses varying from one another by not more than 30 per cent. of the average dose.

4. These three statements may be epitomized by saying that an analysis of 48 courses of administration of digitalis preparations showed that, irrespective of the activity of the preparation used and without relation to the presence or absence of auricular fibrillation, approximately half of the doses fell within 15 per cent. of the average established in terms of the activity of the drug in fractions of a cat unit per pound of body weight of the patient.

The full therapeutic or minor toxic doses of these same preparations, as shown in Tables 4 and 5 for fibrillating and non-fibrillating cases, respectively, have been analyzed in the same way and the results are so closely similar to those just reported for the combined table that it is not necessary to detail them. The remaining average doses for any of the preparations employed, and for cases with or without auricular fibrillation, and for all of the cases together, are set forth in the three tables—4, 5 and 6—and further discussion of them seems unnecessary in this place. They are there for reference, and I shall leave them without further mention except in connection with the subsequent discussion of certain other aspects of our problem.

5. If the therapeutic doses for both tinctures and infusions be taken together as representing the dose of digitalis, the average of the thirty-three courses of administration is 0.146 cat unit per pound of body weight. This will be regarded as the established average dose for digitalis inasmuch as the infusions and tinctures give practically identical figures.

THE MAXIMUM DOSES BORNE WITHOUT DANGEROUS EFFECTS

In any determination of the dose of such a drug as one of the digitalis bodies, in which the administration of sufficient amounts is often a matter of life or death, it would not be enough to fix a dose which could usually be expected to produce therapeutic effects, and it is quite as essential to have some idea of the maximum amount which a patient may be expected to take without suffering injury. These records provide some information along this line, for in some instances the drug was pushed to the point of appearance of some symptom of mild toxic

action when the therapeutic dose had been borne without such manifestation.

In all there were nine instances in which the tincture of digitalis was given in amounts in excess of the full therapeutic dose. Among these nine cases there were three in which at least 50 per cent. more than the average full therapeutic dose was given; two in which approximately 50 per cent. more than this dose was given without so much as the production of emesis, and there was one patient who took nearly 300 per cent. of the average therapeutic dose without the least evidence of any toxic action whatever (Case 17 in Table 3). In none of these cases was there any evident detrimental effect from the digitalis.

In three cases the infusion of digitalis was also given in amounts exceeding the requirements for the production of full therapeutic effects, and one of these took nearly double the average dose without manifesting so much as nausea.

Finally, there were five cases in which the therapeutic dose of digitoxin was similarly exceeded, in one the dose was nearly double, in another more than 1⅔ times the average. In none of the five cases were the excessive doses provocative of more annoying symptoms than nausea and vomiting.

Among all seventeen instances in which one or another preparation of digitalis had been taken in excess of the full therapeutic dose, there was only one instance in which the largest dose borne fell as low as the average therapeutic dose, and in this single case the dose was only 18.5 per cent. below the average.

It is obvious from the preceding discussion that, were we to take the calculated average therapeutic dose as our guide in the administration of digitalis we would run little or no risk of administering a dose which might in any way threaten life.

INFLUENCE OF THE ACTIVITY OF THE PREPARATION OF DIGITALIS ON THE DOSE FOR MAN

It must already be apparent to the reader that the activity of the digitalis leaf from which either a tincture or an infusion is made has no influence on the dose of the resulting preparation if this is measused in terms of the cat unit of activity. Of course the activity of the preparation does have a marked influence on the gross measure of the amount of the preparation which must be given, for either the single doses or the total dose required, when measured in cubic centimeters or in minims or drams.

INFLUENCE OF THE CARDIAC CONDITION ON THE DOSE IN TERMS OF FRACTIONS OF A CAT UNIT PER POUND

Attention has already been called to the fact that the dose required for the production of the full therapeutic effects is essentially the same for any given preparation irrespective of the presence or absence of auricular fibrillation. Throughout this entire series of observations there is no evidence of any relation between the dose required for the production of a given effect and the cardiac condition of the patient. Even in cases in which the heart was not primarily at fault the doses fell within the usual limits established for those in which the heart was the essential offender. Thus, in Table 3, Case 1, the patient showed scarcely any failure of cardiac compensation, and had an old standing mitral leak with associated stenosis apparently of slight grade; the patient in Case 3 had chronic pulmonary tuberculosis and tuberculous peritonitis; the patient in Case 4 was primarily suffering from chronic nephritis; in Case 15 the patient had both chronic nephritis and diabetes mellitus. In the same table the patient in Case 23 was suffering mainly from overwork and mechanical strain of his heart; Patient 31 had chronic interstitial nephritis and mitral stenosis and insufficiency; Patient 32 had chronic cirrhosis of the liver and chronic myocarditis; and Patient 33 chronic syphilitic aortitis.

INFLUENCE OF SEX AND AGE ON THE DOSE

The total number of courses of administration of digitalis (exclusive of the pure principles) amounted to 33, as already stated, and the average therapeutic dose established from these observations is 0.146 cat unit per pound. Nine female patients gave an average dose of 0.167 cat unit per pound, and 24 male patients 0.138 cat unit. The average dose for the females was only 14 per cent. above that for all of the cases, and that for the males only 6 per cent. below. It seems probable that a larger number of cases in both groups would have brought their respective averages even closer to the average for all cases. These facts are presented for what they may be worth.

Analyzing these 33 courses of administration of digitalis from the point of view of age, excluding the two children who required 0.104 and 0.130 cat unit, respectively, the difference in dose due to age is found to be slight. The average dose for patients over 40 years of age is only 5 per cent. above, and that for the patients of 40 or younger only 13 per cent. under the average for all cases. It seems, therefore, that neither age nor sex plays any important part in influencing the dose of digitalis, when this is measured in terms of the cat unit of activity of the drug per pound of the patient's weight.

INFLUENCE OF SIZE AND STATURE ON THE DOSE

Excluding from consideration in this connection the two children, the 31 cases receiving digitalis are divisible into three groups from the point of view of size and stature of the patients. Inasmuch as there was no very obese patient in the entire series, the body weight is a good index of general stature. A group of five patients in which the weight lay between 100 and 125 pounds gave an average therapeutic dose of 0.142 cat unit per pound. Twenty patients ranging from 126 to 150 pounds in weight gave an average dose of 0.141 cat unit per pound. And six patients in which the weight was more than 150 pounds showed an average dose of 0.176 cat unit. Among the six heavy patients only two responded fully to doses less than the established average for all cases. Although the higher figure for the group of heavier patients may be rather more than a mere coincidence, more extended observation is required before any definite statement can be made on this point.

All of the weights stated in the tables were the lowest recorded for each patient, and always represented as nearly as possible the actual weight of the individual after all edema had disappeared. When it was impossible to remove the edema, an allowance was made for the estimated weight of the fluid present, this allowance being based on the actual losses which occurred in other patients. If such an allowance is not made the estimated required dose will usually be decidedly too high. Adipose tissue has almost the same significance as water so far as its influence on the functions of the body are concerned (excepting heat conservation) and it is probable that a similar allowance would have to be made in the calculation of the dose required for a very obese person. This is supported by Hatcher's observation[9] that very fat animals are more susceptible to digitalis in proportion to their weight than are ordinary ones.

RATE OF ABSORPTION OF DIGITALIS AND DIGITOXIN

Most of the earlier patients received the drug in much smaller single doses than I later learned to give, so that the time before the onset of the action of the drug might be expected to have averaged longer among these than among those who received the larger individual doses. The cases have therefore been divided into two groups and Table 7 gives the results of their analysis.

The table shows the marked effect of the size of the individual dose on the time that must elapse before the actions of digitalis or digitoxin can manifest themselves, and it is obvious that if we desire to induce effects rapidly by oral administration we must resort to doses as large as can safely be borne. Cushny[4] has said, "One great limita-

tion in the use of digitalis is caused by the slowness with which its action is elicited. Rarely is any distinct change to be seen before the fourth day of treatment, and this precludes its use in the most acute cases." This is the prevalent idea among clinicians and to be found in the text-books. That it is incorrect seems so obvious that it is a matter of surprise to find that very few have taken exception to it. One clinical investigator[5] has recently said that by the use of large doses, up to two drams of the tincture of digitalis (B. P.) daily, nausea and beneficial cardiac effect could be produced in from thirty-six to forty-eight hours. To this I can add from my own experience that I have not infrequently succeeded in producing full therapeutic effects from the oral administration of digitalis or digitoxin in from twelve to eighteen hours after the first dose was given, and could

TABLE 7.—Showing Average Time from First Dose to Onset of Effects of Digitalis and Digitoxin

	Individual Doses	Earliest Effects		Full Therapeutic Effects	
		Number of Cases	Average Time in Hours	Number of Cases	Average Time in Hours
Digitalis	Small	8	38	19	70
	Large	16	13	12	28
Digitoxin	Small	5	42	7	84
	Large	8	15	8	33

induce these effects in the majority of cases in about twenty-four hours by the use of large doses adjusted to the patient's needs on the basis of the cat unit of activity of the preparation and the weight of the patient. On this basis I have given single doses of the tincture of digitalis ranging from 8 to 15 c.c.; of the infusion, an initial dose of 50 c.c.; and have used single doses of crystalline digitoxin of from 1.0 to 1.75 mg. As shown in Table 7, the average time for the development of the full therapeutic effect after digitoxin has been given in large individual doses is less than thirty-six hours, and the full effects have been secured in as little as twenty hours on more than one occasion. It seems probable, with a better understanding of the dosage of this principle, that its full action can be secured quite as rapidly as that from the galenical preparations of digitalis. The statements in the literature concerning the phenomenal toxicity of digitoxin and its peculiar slow development of action seem to have been somewhat misleading.

The personal experience recorded by Koppe[13] has been widely cited as indicative of the great toxicity of digitoxin and its slow absorption. Koppe took 0.5 mg. without effect, and twenty-three hours later took 1.0 mg. This was followed by some slight evidence of its action, but this was not sufficiently noticeable to prevent his taking a third dose four days after the second, this time of 2.0 mg. Some hours after this last dose he became ill and the symptoms he described have been taken as indicative of very serious poisoning by the drug. He took in all only 3.5 mg. in five days, and in the light of the doses recorded in this present study it seems probable that we must seek some explanation other than that of digitoxin action alone to account for Koppe's symptoms. The dose that he took in a period of five days certainly falls well within the limits here established for the therapeutic dose for an average adult when given in from two to four days' time.

It should be emphasized, however, that the use of such large doses of either digitalis or digitoxin is too dangerous for general practice and is possible only when the patient can be under almost constant observation and when the action of the drug can be observed both clinically and by means of the polygraph or electrocardiograph. One must also be certain that the patient has not been under digitalis administration for several weeks before being treated.

The present observations, which show that the full therapeutic effects of digitoxin can be obtained in from twenty to forty hours, with an average required time of thirty-three hours, when moderately large doses are given, stand in direct contradiction of Fraenkel's statements.[14] He says that sixty hours elapse after subcutaneous injection before the effect on the pulse appears; that it is at least twenty-four hours before slowing is demonstrable after either therapeutic or toxic doses; and that even after several times the fatal dose a cat will not die in less than from six to twelve hours. His contention is that digitoxin is slowly taken up by the heart even after it has been absorbed into the circulation. Not only do the present observations on man oppose this view, but Hatcher[15] has shown this to be wholly incorrect for the cat, on which animal Fraenkel's experiments were made.

From the preceding facts, therefore, it seems obvious that both digitalis and digitoxin are usually absorbed quite rapidly from the alimentary canal of man, statements to the contrary notwithstanding.

13. Koppe, R.: Untersuchungen ueber pharmakologischen Wirkungen des Digitoxins, Digitalins, und Digitaleins, Arch. f. exper. Path. u. Pharm., 1875, iii, 274.

14. Fraenkel, A.: Vergleichende Untersuchungen ueber die Kumulative Wirkung der Digitaliskoerper, Arch. f. exper. Path. u. Pharm., 1904, li, 84.

15. Hatcher, Robert A.: The Persistence of Action of the Digitalins, The Archives Int. Med., 1912, x, 268.

THE COMPARATIVE ABSORPTION OF DIGITALIS AND DIGITOXIN FROM THE ALIMENTARY CANAL OF MAN

Aside from the general knowledge that the oral administration of these drugs usually produces the effects associated with their absorption we have relatively meager information on this important question, so far as it concerns man. I have already cited the observations of Bailey,[7] Hatcher and Bailey,[8] and Hatcher[9] that ouabain, strophanthus and the strophanthins are absorbed from the alimentary canal of either man or animals in a very variable manner, but for the most part both slowly and incompletely. From repeated observations which Dr. Hatcher and I have made in the laboratory we have come to the conclusion that this is also true of many other digitalis bodies, among which the true digitalin of Kiliani is to be included.

The dose of true digitalin is variously stated by different observers, and New and Nonofficial Remedies for 1914 says that some authorities give the same doses as for crystalline digitoxin (0.25 mg. or 1/250 grain) while others give much larger doses. Klingenberg[16] could not secure any very marked effects from doses up to 15 mg. daily, and I have given as much as 48 mg. daily to a patient for more than four days before the full therapeutic action was secured. On the other hand only about one-sixth of this amount proved adequate for another patient. In terms of cat units per pound the largest dose required for full therapeutic action was 7.6 times as great as that of digitoxin. In view of this apparent lack of absorption I abandoned the use of true digitalin and have made no attempt to establish its dose.

My experience with digitalein is rather similar to that just recorded, and as this is not a pure principle and is not of uniform composition I have omitted it from these studies. We may, therefore, confine our attention to digitalis as a whole and to digitoxin.

It is obvious from the fact that it has been possible to fix a dose for each of these preparations, from which the range of variation is not very great in spite of the many complicating factors, that both of them are absorbed from the alimentary canal of man in a fairly uniform manner. Both seem also to be absorbed with considerable rapidity, as is demonstrated by the fact that the full therapeutic effects can be induced within comparatively few hours after the administration of the first dose. When the administration of either drug is stopped at once after the appearance of minor toxic symptoms or the evidences of full therapeutic action there is little or no increase in either group of phenomena. In fact, if the administration is checked

16. Klingenberg: Ueber die klinische Bedeutung des Digitalinum verum, Arch. f. exper. Path. u. Pharm., 1894, xxxiii, 353.

at the appearance of nausea, vomiting often fails to appear. Both of these facts indicate that the absorption is completed in a relatively short time. Prompt and efficient absorption seems also to take place even in the face of considerable abnormality of the alimentary canal, for patients manifesting evidence of marked congestion of this region, resulting even in repeated vomiting, respond quite as promptly and to the same doses as do those who are apparently free from disturbance. A case in point is Case 4, under Report of Illustrative Cases.

The establishment of the fact that both digitalis and digitoxin are usually promptly and fairly uniformly absorbed in man does not prove, however, that they are absorbed with equal rapidity or to an equal extent and the following seems to show that such is actually not the case.

Hatcher[9] has shown that the several digitalis bodies are mutually and quantitatively synergistic in their actions on the heart in the cat and other animals. If the fatal vein dose be established for digitalis and digitoxin, respectively, in the cat and then each of several other cats be given 50 per cent. of the fatal dose of one of these, it will require an amount of the other to cause death which is 50 per cent. of its fatal dose. In other words 50 per cent. of the fatal vein doses of each of these two preparations is equivalent to 100 per cent. of the fatal dose of either. If, therefore, digitoxin and digitalis were absorbed to an equal extent from the alimentary canal of man the doses of these two, in terms of fractions of a cat unit per pound, should be the same for the production of the same degree of action. The results of the present observations show that such is not the case. Thus, the therapeutic dose of digitalis is 0.146 cat unit per pound while that for digitoxin is only 0.066 cat unit. In other words, in terms of activity it requires 2.21 times as much digitalis as of digitoxin to produce the same effect when both are given orally to man.

The leaf from which Tincture 97 was made was assayed by C. E. Vanderkleed and found to contain 0.31 per cent. of digitoxin. Eleven patients weighing 1,543 pounds total required 21,700 mg. of this leaf for the production of full therapeutic effects, giving an average of 14 mg. per pound. This is equivalent to 0.0434 mg. of digitoxin per pound. Fifteen patients required an average of 0.023 mg. of crystalline digitoxin per pound for the production of a similar effect. On the basis of weight of digitoxin these figures show that when given in the form of the tincture of digitalis 1.88 times as much digitoxin was required for the production of a given effect as when the pure digitoxin itself was given. We may account for this fact in one of two ways. Either on the ground that there must be some substance present in digitalis and soluble in both alcohol and water (tincture and infusion) which

acts to delay the absorption of the digitoxin present. Or by supposing that the digitoxin in the leaf and its galenical preparations is present either in a different form from that in which we know it after isolation, or is bound to some other substance in a way which permits of the liberation of only a portion of it for absorption. This latter would seem to be the more plausible explanation in view of the complexity of plant constituents.

This fact also serves to confirm the belief that the chemical assay of digitalis, in which the digitoxin content is determined, does not provide results which can be translated into doses for man, and it gives a rational explanation for this deficiency of the method.

METHODS OF STANDARDIZATION COMPARED WITH REFERENCE TO THEIR APPLICABILITY TO THE DETERMINATION OF DOSAGE FOR MAN

In another paper[17] I have discussed the relative merits of the several biologic methods of standardizing digitalis preparations, and there made the statement that one of the desiderata of any method should be that, "The results of the evaluations should be more or less fully transferable to man." At the conclusion of the paper I expressed the opinion that the cat method of Hatcher attained this end. This opinion was based mainly on critical analysis and was little supported by clinical observation, except in a few details. The results of the present work now seem to have established this as a fact. There are, however, other methods of standardization, at least one of which is quite widely accepted—the one-hour frog method. Dr. Hatcher and I have contended that by this, or any other method in which absorption of the drug is an essential feature, it would be impossible to compare the activities of two different samples of a complex substance like digitalis in a way which would give results transferable to man. I am now in a position to offer confirmation for this contention.

The leaves from which Tinctures 97 and 150 were prepared were found by Vanderkleed to contain 0.31 and 0.27 per cent. of digitoxin, respectively. In terms of digitoxin, therefore, the former was 1.14 times as active as the latter. The frog test showed Tincture 97 to be 1.19 times as active as Tincture 150. On the cat Tincture 97 proved to be 1.54 times as active as Tincture 150, and on man, based on the number of milligrams of the leaf per pound of body weight which was required to produce therapeutic effects, the former was 1.71 times the latter in activity. The accompanying tabular presentation (Table 8) shows these relations more clearly.

17. Eggleston, Cary: Biological Standardization of the Digitalis Bodies by the Cat Method of Hatcher, Am. Jour. Pharm., 1913, lxxxv, 99.

Here the chemical determination and the one-hour frog tests gave similar ratios of activity for the two specimens, but between these ratios and those determined on the cat and on man there is considerable divergence. On the other hand, the ratio of activity established by the cat is very nearly the same as that found to hold for man. It must be borne in mind that in the case of the dose of Tincture 150 we have only three observations, the average of which happens to be higher than that for digitalis in general, as founded on a large series. If there had been a larger number of cases treated with this tincture the average dose would almost certainly have approached that for digitalis, namely, 0.146 cat unit per pound. Had this been the case, the ratio of activity between Tinctures 97 and 150, as based on the number of milligrams of the leaf taken per pound, would have fallen to from 1 to 1.57, almost exactly that determined on the cat. The facts, discarding this suggested correction, certainly show that the ratio

TABLE 8.—Relative Activity of Two Tinctures of Digitalis Prepared from Leaves Containing Varying Amounts of Digitoxin

In Terms of Acidity as Determined	Tincture 97		Tincture 150
Chemically	1 part	equals	1.14 parts
On frogs	1 part	equals	1.19 parts
On cats	1 part	equals	1.54 parts
On man	1 part	equals	1.71 parts

of activity determined on the cat is much more nearly that for man in the case of digitalis, than is the ratio similarly determined with the same specimens by either the frog or the chemical methods of standardization.

When it comes to the comparison of the relative activity of a sample digitalis with that of digitoxin all methods fail to give results which are transferable to man on account of the peculiarity of the absorption of digitoxin from the galenical preparations of the leaf. Thus, by the frog, digitoxin would be 247 times as active as the leaf of Tincture 97 and 277 times as active by the cat test, but it proved to have been 606 times as active on man. This failure of the several methods was anticipated, and we have held from our animal experiments that such was necessarily the case. This in no way conflicts with the contention that the cat method is the best suited for the determination of the relative activity of different digitalis bodies after they have gained entrance into the circulation.

Before passing to the summary and conclusions to be drawn from these observations I should like to record a method of practical application of their results which naturally suggests itself.

PRACTICAL APPLICATION

It would be desirable to prescribe a preparation of digitalis which had been standardized by the cat method, but if this was not obtainable one could proceed on the basis that a high grade leaf had an average cat unit strength of 100 mg., and the cat unit should then be used as the basis for the calculation of the probable required dose. In the case of an average first-class tincture 0.145 c.c. could be taken as the average therapeutic dose for each pound of the patient's body weight. On the basis of the patient's actual or estimated weight the total amount which would probably be required should be calculated and this quantity could then be divided into single and daily doses according to the rapidity with which it was desired to induce the full therapeutic effects. If after the total calculated amount had been taken the patient failed to show the full therapeutic effect, or some minor toxic action indicating that enough had been given, the administration should be continued in small repeated doses until one or the other of these evidences called for its withdrawal.

In this way it is possible to give a third to half of the total calculated therapeutic dose at a single administration, to follow this in from four to six hours with a quarter to a third of the total dose, and to give the remainder in a few doses of smaller size at intervals of from four to six hours. By this plan of administration the full effects can be secured in from twelve to thirty-six hours in the majority of cases.

The administration of half of the total dose may call for the giving of from 5 to 15 c.c. of the tincture at once, and it might be feared that such a large dose might cause gastric irritation and nausea or vomiting. I have given such doses repeatedly since the completion of the greater portion of this work and have never seen the least disturbance of any kind arising as a consequence. This is due to the fact that the nausea and vomiting following the administration of the digitalis bodies is of central origin and results only after the absorption of a sufficient quantity of the drug into the circulation. Dr. Hatcher and I have previously shown this to be true for both man and animals.[18]

The same plan could be carried out with digitoxin, which is approximately of uniform activity, the average dose per pound for man being 0.023 mg. It would be better to employ this preparation in the form of tablet triturates, as fairly strong alcohol is required for its

18. Hatcher, Robert A., and Eggleston, Cary: The Emetic Action of the Digitalis Bodies, Jour. Pharm. and Exper. Therap., 1912, iv, 113. Eggleston, Cary, and Hatcher, Robert A.: The Emetic Action of the Digitalis Bodies, Jour. Am. Med. Assn., 1913, lx, 499. Eggleston, Cary: Clinical Observations on the Emetic Action of the Digitalis Bodies, ibid., 1913, lxi, 757.

solution and the evaporation of some of the solvent might lead to such a concentration of the solution that a dangerous miscalculation might result, or the substance might become partly precipitated.

It should be reiterated in this place that the use of such large doses of either digitalis or digitoxin as are here mentioned is not a safe procedure unless the patient can be under nearly constant observation and unless the effects of the treatment can be graphically recorded at frequent intervals. This practically limits such procedures to hospital practice and to those well versed in the significance of polygraphic and electrocardiographic records.

An interesting bit of evidence may be introduced here which tends to confirm the correctness of the preceding statements of average doses. It might be added that this confirmation was quite unexpected. In a paper on the emetic action of digitalis[18] I reported a considerable number of courses of administration of digitalis which were carried to the production of nausea or vomiting. In all of these cases the daily doses given were rather small and the administration was continued over a considerable period of time, so that the doses taken were almost certainly a little in excess of the minimum which would have been needed had larger daily doses been given. The doses taken were known in terms of grams of digitalis leaf for each of the cases. In 68 first courses of administration the average dose per patient was 3,666 mg. of leaf. On the basis of 150 pounds as the average weight of an adult this would have amounted to an average dose of 30.5 mg. of leaf per pound of body weight. We have tested a large number of different samples of digitalis in the laboratory and have established 120 mg. as the cat unit of the average specimen of digitalis of commerce. With this as the activity the average emetic dose for the 68 cases would be 0.203 cat unit per pound. The 14 instances of nausea or vomiting in Tables 1 and 3 of this article give an average emetic dose of 0.187 cat unit per pound. These two figures are very similar in view of the difference in the methods of observation employed in the two series of cases providing them.

SUMMARY

1. The confused state of our knowledge of the dosage of the digitalis bodies has been set forth.

2. A series of observations has been reported and the results subjected to analysis from several points of view for the purpose of gaining some information on the subject of the dosage of these bodies for oral administration to man, and from these observations and analyses the following conclusions and deductions can be offered.

CONCLUSIONS AND DEDUCTIONS

1. The cat method of standardization of digitalis yields results on which the dose for man can be based.

2. The average therapeutic dose of digitalis, given orally to man in the form of the tincture or infusion, is 0.146 cat unit or about 0.146 c.c. of an average high-grade tincture per pound of body weight, as established by thirty-three observations.

3. Fifteen observations have established 0.066 cat unit, or 0.023 mg., per pound as the average therapeutic dose of crystalline digitoxin.

4. In approximately half of a total of 48 courses of administration of either digitalis or digitoxin, full therapeutic effects were secured with doses falling within 15 per cent. above or below the average dose.

5. Doses considerably larger than the average were taken in 17 instances without the production of more than mild toxic symptoms.

6. The activity of the preparation of digitalis has no material influence on the dose required in terms of cat units.

7. Age, sex, and cardiac condition do not seem to influence the size of the dose required.

8. Both digitalis and digitoxin are probably rapidly and fairly uniformly absorbed from the alimentary canal of man, but digitalis is less completely absorbed than is digitoxin.

9. Strophanthus, the strophanthins, ouabain, true digitalin, and some other digitalis substances are poorly or irregularly absorbed when given by mouth to man or to the higher animals and are unsuited for therapeutic use in this way.

I wish to acknowledge the help I have derived in the course of this work from many profitable discussions with Dr. Hatcher. Dr. C. E. Vanderkleed's cooperation in supplying me with digitalis leaves, the digitoxin content of which he had determined, is also much appreciated. I have, finally, to thank the attending and house staffs of the Second Medical Divisions of Bellevue and of City Hospitals, respectively, for their cordial cooperation in providing me with the cases for this study. I should like to mention, in particular, Dr. E. P. Shelby of the City Hospital and Dr. Warren Coleman of Bellevue Hospital attending staffs without whose generous aid I should not have been able to have made these observations for want of sufficient clinical material.

414 East Twenty-Sixth Street.

THE DIAGNOSTIC VALUE OF URIC ACID DETERMINATIONS IN BLOOD*

PROF. OTTO FOLIN AND W. DENIS, PH.D.
BOSTON

Ever since the description by Garrod in 1860 of his so-called thread test for uric acid in gouty blood, the finding of an excess of uric acid in the blood of patients suffering from joint diseases has been considered to point toward a diagnosis of gout. Within recent years Brugsch and Schittenhelm[1] have stated that in normal human blood uric acid is absent. Its presence they consider as pointing to the existence of gout. This view has also been accepted by Gudzent[2] who recommends his dialysis method for the detection of uric acid in blood as being of aid in the differential diagnosis of joint conditions.

That Brugsch still believes in the diagnostic value of uric acid tests is shown by the recent appearance of a paper from his laboratory[3] describing a clinical method for the determination of this substance in blood. According to this procedure a few drops of serum are treated in a special test tube by means of our phosphotungstic acid reagent. Sodium carbonate is then added and the blue color obtained is read against a permanent standard. Coming from Brugsch such a method will doubtless receive considerable attention, and will lead to much useless and misleading work. All blood contains varying amounts of phenols, which also give a blue color with our phosphotungstic acid reagent; indeed, in some cases these bodies will give from two to three times the amount of color produced by the uric acid present in the blood. In our method the uric acid is precipitated and thus separated from the phenols. We realize that the determination of uric acid in blood by our method is still probably outside the range of most clinical laboratories, but we are convinced that all "short cut" methods so far proposed are bound to lead to grossly misleading results.

By means of our technic we were able to show about three years ago that 100 c.c. of normal human blood contains 1.5 to 2.5 milligrams of uric acid, and that in gout, lead poisoning, leukemia, and in same cases of nephritis this amount was greatly increased.

---

* Submitted for publication March 3, 1915.

* From the Chemical Laboratories of the Massachusetts General Hospital and Harvard Medical School.

1. Brugsch and Schittenhelm: Ztschr. f. exper. Path. u. Therap., 1907, iv, 438, 446, 480, 532, 538, 551.
2. Gudzent: Deutsch. med. Wchnschr, 1912, xxxviii, 603.
3. Brugsch and Krysteller: Deutsch. med. Wchnschr., 1914, xl, 746.

From the results of our quantitative uric acid determinations in normal human blood it became clear at once that the various qualitative tests for uric acid, from Garrod's naive thread test down to the elaborate recent procedures of Schittenhelm and of Gudzent are all equally treacherous and useless for differential diagnostic purposes. The value of exact quantitative determinations of the uric acid for such purposes seemed, on the other hand, to rest on a sound foundation of facts. The accumulation of relatively immense urate deposits in the joints of persons suffering from gout would seem to be almost conclusive proof that the circulating fluids of such patients must be particularly rich in urates in comparison with the blood of those in whom such deposits of urates never occur. The blood of normal persons has, however, been found to carry far more uric acid (1-2.5 mg. per 100 c.c.) than was formerly suspected, and the difference in the uric acid content of such normal blood and the blood of those suffering from gout is materially smaller than earlier investigators realized. Since it is by no means excluded that the blood in diseases other than gout may not, occasionally at least, carry more than the normal amount of uric acid, the diagnosis of gout by means of uric acid determinations is by no means so simple or certain that numerous and serious blunders will not occur. In the course of several hundred uric acid determinations made on many different kinds of human blood during the past three years we have become convinced that even exact quantitative uric acid determinations are not by themselves an adequate protection against frequent mistakes in the differential diagnosis of gout and other joint diseases.

In our earlier work we were struck by the fact that there is apparently no constant relationship between the amount of uric acid and the amount of nonprotein nitrogen in human blood. The variations found are very much greater than the corresponding variations (between the uric acid and the total nitrogen) met with in urine on a given, definite diet, and seemed to be due to a selective excretory activity on part of the kidneys. In respect of these two factors, the uric acid and the nonprotein nitrogen, there manifestly can be four distinctly different classes of human blood.

1. Blood in which both uric acid and nonprotein nitrogen are present in normal amounts.

2. Blood in which with normal amounts of uric acid we have greatly increased amounts of nonprotein nitrogen.

3. Blood giving abnormally high uric acid values with normal amounts of nonprotein nitrogen.

4. Blood in which abnormally large amounts of both uric acid and nonprotein nitrogen are present.

Examples of each of these classes are given in the accompanying tables.

TABLE 1, CLASS 1

NORMAL URIC ACID AND NORMAL NONPROTEIN NITROGEN

No.	Diagnosis	Milligrams per 100 c.c. Nonprotein Nitrogen	Uric Acid
	Normal man	32	2.0
	Normal man	24	2.0
107	Alcoholic gastritis	34	2.0
147	Cystinuria	28	1.4
9	Diabetes insipidus	25	1.5
144	Cardiorenal disease, arteriosclerosis	26	2.8
P 6	Insanity	32	2.4
P 6	Insanity	24	2.0
P 6	Insanity	28	2.0
P 6	Insanity	28	2.4
S 8	Arteriosclerosis	30	2.5
S 9	Chronic interstitial nephritis	27	2.6
S10	Chronic interstitial nephritis	30	2.5

TABLE 2, CLASS 2

NORMAL URIC ACID AND HIGH NONPROTEIN NITROGEN

No.	Diagnosis	Milligrams per 100 c.c. Nonprotein Nitrogen	Uric Acid
10	Nephritis, prostatectomy	110	2.3
R16	Infectious arthritis (purin free diet for two days)	89	1.8
R14	Bone tuberculosis (purin free diet for two days)	102	1.6
R18	Acute rheumatic fever (purin free diet for two days)	104	1.7
R19	Acute rheumatic fever, pericarditis	100	1.6
R36	Mitral stenosis (purin free diet)	60	2.2
T1	Arthritis deformans	54	1.2
T9	Arthritis deformans	50	2.0
T20	Infectious arthritis	80	2.3
T26	Arthritis	50	2.0
58	Acute infectious arthritis (purin free diet)	80	1.9

TABLE 3, CLASS 3

HIGH URIC ACID AND NORMAL NON-PROTEIN NITROGEN *

No.	Diagnosis	Milligrams per 100 c.c. Nonprotein Nitrogen	Uric Acid
1	Typical gouty attacks for past seven years	25	3.8
3	Arteriosclerosis. Acute gout, first attack	40	3.4
4	Alcoholic gastritis and gout	40	3.5
5	Gout, many tophi	30	4.4
6	Typical gout, last attack two years ago	28	5.2
12	Has passed several vesical calculi consisting of pure uric acid	30	5.2
L1	Typical gout in both great toes	36	5.4
D3	Acute gout, many tophi, arteriosclerosis	32	3.1
L6	Acute gout, many tophi, large urate ulcer	42	5.1
L	Normal man, many members of whose family have been gouty	29	4.0
49	Swollen joints, probable gout	28	5.0
106	Swollen and painful joints, probably gout; no tophi	30	3.3

* Purin-free diets for at least two days before blood was taken.

TABLE 4, CLASS 4

HIGH URIC ACID AND HIGH NONPROTEIN NITROGEN

No.	Diagnosis	Milligrams per 100 c.c. Nonprotein Nitrogen	Uric Acid
82	Uremia	288	9.5
97	Uremia	284	6.6
28	Uremia	125	10.0
76	Uremia	228	7.5
R16	Arthritis deformas (purin free diet for two days)	104	3.8
R32	Weak heart, edema of lungs, hypertrophic arthritis	90	5.0
38C	Pneumonia	72	5.0
78C	Cardiorenal case	326	4.4
77C	Chronic nephritis	148	6.5
29C	Nephritis following eclampsia	100	4.1
T3	Arthritis deformans	62	7.0
T22	Acute gonorrheal arthritis	124	3.3
T32	Arthritis deformans	59	3.6
L4	Acute gout, tophi, chronic interstitial nephritis (purin free diet)	60	5.7

The last three tables represent three chemically different kinds of abnormal blood.

Gout, as will be seen from Table 3, is characterized by abnormally high uric acid content of the blood *without* any abnormally high accumulation of other nitrogenous waste products (nonprotein nitrogen). Exceptions may of course occur; gouty patients may also have nephritis. Nephritis in the gouty is usually of the arteriosclerotic type, a type which according to our experience is not accompanied by an excessive accumulation of nonprotein nitrogen in the blood (except in the terminal stages of the disease). This rule is, however, unfortunately not without exceptions. The last case quoted in Table 4, of gout and interstitial nephritis, showed a high nonprotein nitrogen (60 mg.) as well as a very high uric acid (5.7 mg.).

From an inspection of Table 3 and by comparing the figures and diagnoses there given with the figures and diagnoses of Tables 2 and 4, it will be seen that to be of material help in the differential diagnosis of gout the uric acid determinations must be accompanied by determinations of the nonprotein nitrogen. In joint diseases other than gout it is by no means uncommon to find uric acid values nearly if not quite so high as in gout. In arthritis, however, a high nonprotein nitrogen is even more frequent than a high uric acid. Indeed, in this respect the blood in arthritis frequently resembles the blood of glomerular nephritis.

In making use of blood analysis to decide whether a given doubtful case of joint disease is gout or arthritis, it is therefore absolutely necessary to determine the nonprotein nitrogen (or at least the urea) in the blood as well as the uric acid. And before the blood is drawn for such analyses it is indispensable that the patient should have been

on a purin-free diet for at least two days. The level of the protein metabolism should also be ascertained by means of a nitrogen determination in the twenty-four hour urine passed during the last day of the experiment.

SUMMARY

1. In gout the blood is almost invariably abnormally high in uric acid, while the other waste products represented in the nonprotein nitrogen of the blood are usually within the normal limits. In arthritis also the blood is not infrequently abnormally high in uric acid, but most such cases have abnormally high nonprotein nitrogen as well.

2. Neither qualitative tests for uric acid in the blood nor quantitative determinations of the uric acid alone can be depended on in the differential diagnosis of gout and other joint diseases.

3. For a differential diagnosis in doubtful cases of gout or arthritis by means of blood analyses the patient must be put on a purin free diet and uric acid determinatnons must be accompanied by determinations of the nonprotein nitrogen (or urea).

Harvard Medical School.

A CASE OF OPEN DUCTUS ARTERIOSUS (BOTALLI), WITH NECROPSY *

JAMES L. STODDARD, M.D.
BOSTON

A case of open ductus Botalli in a girl of 17, with necropsy, occurring at the Peter Bent Brigham Hospital in the last year, was carefully observed on Dr. Christian's service in the hospital for fifteen days, and notes were obtained of a previous examination four years before entrance. Diagnosis of the lesion was made during life. The completeness of the record of this case gives it that value as an aid to the diagnosis of the lesion which is common to all carefully observed single cases. The purpose of this paper is to present and comment upon this case, to summarize the recent uncollected cases, and to consider such points as these data seem to throw light on, but to make no attempt to repeat the general discussion of the subject so fully presented by Hochsinger,[1] Wells,[2] Abbott,[3] and Goodman.[4]

In 1908 H. G. Wells collected all the uncomplicated cases of open ductus Botalli and found a total of forty-one, only twenty of which were in adults. Maude Abbott, in the same year, found thirty-seven, only nineteen of which had clinical histories, and three of these latter cases were unconfirmed by necropsy. E. H. Goodman, in 1910, determined the percentage occurrence of symptoms in thirty-four necropsied cases. It is not possible to tell from these papers how many cases confirmed by necropsy and carefully recorded clinically have occurred in adults, but the number is evidently extremely small.

Dr. Abbott's and Dr. Goodman's analyses of the cases reported are so complete that I have used them in my summary. I have carefully searched the literature for cases occurring since the publication of these papers, and have found twenty-two in all. In only five was the patient over 1 year old; one of these cases was complicated by slight aortic and mitral stenosis, and one by mitral stenosis with acute vegetations; none

* Submitted for publication Jan. 29, 1915.
* From the Pathological Laboratory of the Peter Bent Brigham Hospital.

1. Hochsinger, K.: Zur Diagnose der Persistenz des Botallischen Ganges und der Erweiterung der Lungenarterie, Wiener Klinik, 1907, xxxiii, 311.
2. Wells, H. G.: Persistent Patency of the Ductus Arteriosus Botalli, Am. Jour. Med. Sc., 1908, cxxxvi, 381.
3. Abbott, M. E.: Congenital Cardiac Disease, Osler's Modern Medicine, 1908, iv, 323.
4. Goodman, E. H.: Report of a Case of Patent Ductus Arteriosus Botalli, with a Study of the Cases Heretofore Published, Univ. Pennsylvania Medical Bulletin, 1910, xxiii, 509.

had other important congenital lesions. Of the infantile cases eleven were complicated by other congenital abnormalities, leaving two uncomplicated infantile cases and two infantile cases in which an absence of other lesions was not noted. The symptomatology and pathology will be discussed after the presentation of the case I have studied.

REPORT OF CASE

Patient.—E. S., aged 17, female, unmarried, of German ancestry. At the age of 7 the patient had measles, and shortly afterwards mumps. Her general health had been good; she was not subject to tonsillitis or colds, and no other infectious diseases had been noted. There was no history of decompensation symptoms such as dyspnea, edema, or precordial pain. There were no symptoms referable to other organs, with the exception of headaches, occurring about once a week.

In March, 1910, at the age of 13, the patient entered the service of Dr. Christian at the Carney Hospital, Boston, and was discharged after thirteen days with the diagnosis of "Congenital heart. Pulmonic stenosis. (?) Open ductus arteriosus (?)."

First Examination.—On physical examination at entrance, the following heart condition was found: Dulness, the upper border at the third rib, the right border 1 cm. to the right of the midsternal line; the left border 3.75 cm. to the left of the midsternal line. A palpable thrill over the pulmonic area extended to the left and downward along the left sternal margin. Over the same area was heard a rough systolic rumble running into diastole. A systolic murmur was heard all over the precordia. The pulmonic second sound was accentuated. The action was regular.

March 16 the thrill was noted to be continuous through systole and diastole, but somewhat stronger in systole, with a late systolic accentuation. At the apex could be felt a short late diastolic and a systolic thrill.

June 19 the patient entered the Peter Bent Brigham Hospital on account of "spots on the legs." There had been no general malaise or other symptoms of illness. Four days before entrance a large black and blue spot had appeared spontaneously on the inner side of the right ankle, and soon after several small reddish spots on both shins. The spots had given no noticeable subjective sensations.

Second Examination.—A rather poorly developed and nourished girl was found, in no apparent discomfort. The only positive points on physical examination were with reference to the heart and extremities.

The heart examination showed: On palpation, an apex impulse in the fifth space, 8 cm. to the left of the midsternal line; over the pulmonic area a marked thrill running throughout the cardiac cycle with greatest intensity during systole; a sharp second sound, distinctly felt, over the pulmonic area. On percussion, dulness with the upper border at the third rib, the right border 2 cm. to the right of the midsternal line, and the left border 10 cm. to the left of the midsternal line in the fifth interspace. On auscultation, at the apex a first sound of good quality, followed by a systolic murmur somewhat rumbling in character, apparently transmitted from the pulmonic area; a normal second sound; a loud rough murmur over the pulmonic area, running throughout the cardiac cycle coincident with the thrill, with the greatest intensity during systole; a blowing diastolic murmur, distinct from the other murmurs, transmitted from the pulmonic area down along the left border of the sternum, becoming slightly musical in the third and fourth interspaces; aortic second not increased; pulmonic second markedly accentuated; the action regular and moderately rapid. The blood pressure was 118 systolic, 70 diastolic.

Extremities: The fingers were clubbed and the nails cyanotic. On the right leg, just above the internal malleolus, was a purplish-black echymosis, about 5 mm. in diameter, moderately tender. Scattered over the anterior surfaces of both shins were a number of small purpuric spots 1 to 3 mm. in diameter.

The temperature was 103.4, pulse 129, respiration 31. The white count was 14,000, and the hemoglobin 65 per cent. A smear showed moderate achromia, polymorphonuclears 78 per cent., small mononuclears 7 per cent., large mononuclears 15 per cent., eosinophils none. The urine had a small trace of albumin. The Wassermann reaction on the blood serum was negative.

The clubbing of the fingers, with some cyanosis, the character and localization of the thrill and murmurs, and the lack of etiology for cardiac disease, suggested a congenital cardiac lesion, either a patent foramen ovale, or a patent ductus arteriosus. A severe active endocarditis superimposed on the congenital defect was indicated by the purpuric spots, fever, anemia, and heavy sweats; although this indicated infection was considered not necessarily localized in the heart.

Further Clinical Notes.—On June 22 examination gave similar heart findings, except that the systolic and diastolic murmurs were noted all over the precordia, and the diastolic murmur seemed of greatest intensity over the aortic area, suggesting a diagnosis of aortic insufficiency in addition to an open ductus arteriosus. The murmur over the pulmonic area is described as a "sawing to and fro murmur" through the whole cycle.

June 23: A high evening temperature, usually reaching 103, continued, but the patient felt comfortable and had no complaints. The purpuric spots had almost faded out.

June 25: Slight cyanosis was noted. Visible pulsations were found in the fifth space, nipple line, in the suprasternal notch, and in the second interspace to the left of the sternum. In the latter spot the impulse was wave-like. The time of maximum intensity was just after the first sound, as judged by apex and aortic pulsations. On percussion no increase of dulness was definite to the right of the sternum, while to the left in the second and third spaces increased dulness was found for a distance from 1 to 2 cm. from the sternum. On auscultation the first sound appeared blurred, and was followed by a low-pitched murmur transmitted into the axilla. The rhythm was a protodiastolic gallop. Otherwise the examination revealed no new facts. Another observer the same day noted a distinct capillary pulse.

June 26: The total diameter of the heart was made out by percussion to be 18.5 cm., the left border being 15 cm. to the left of the midsternal line at the level of the fifth rib, and the right border 3.5 cm. to the right of the midsternal line at the junction of the fourth rib. The apex was most distinct in the fifth space, but could be felt in the sixth. A pistol shot sound was heard in the femoral arteries.

June 27: A distinct systolic thrill at the apex was noted. The diastolic murmur seemed most intense in the third space and the systolic at the apex. A capillary pulse in the fingers and collapsing pulse in the small arteries were plain. The diagnosis at this time was aortic and mitral regurgitation, with possibly a patent ductus arteriosus, and with an acute process on the chronic valvular lesions.

Two blood cultures had showed no growth. Widal reactions were negative. The white count had risen to 34,000. The red count was 2,408,000. Occasional granular casts were found in the urine. The temperature and pulse continued the same course.

The patient's condition now began to grow worse; there was much vomiting, and no food intake. The temperature fell to 99 June 29 and remained about at that level until July 3, when there was an ante-mortem drop.

On July 2 dulness with many fine crackling râles were found at the right base, and the white count was 70,000.

July 3 the systolic murmur was heard in the vessels of the neck, and seemed most intense over the sternum at the level of the third costal cartilage; the diastolic was plainest over the pulmonic area and along the sternum; a capillary pulse, a Corrigan pulse, and pistol shot sounds in the femoral arteries were plainly observed. Cyanosis persisted, and there was dyspnea out of proportion to the temperature.

On July 4, after a comfortable night, the patient sat up in bed suddenly at about 9 o'clock in the morning, with a complaint of palpitation and dyspnea, and quickly became very cyanotic and almost pulseless. The right border of

Fig. 1.—View of the heart from the left showing the vegetations on the aortic valve (*B*), and a probe in the open ductus (*A*). Beneath the probe can be seen the ridge on the aorta.

the heart was found to be 5 cm. (later 6 cm.) to the right of the midsternal line. The liver was felt 6 cm. below the costal margin and was pulsating. The previously felt thrills had almost disappeared, and the murmurs were of almost the same character, but of diminished intensity. A presystolic gallop rhythm was felt at the apex. The lungs appeared as before. The condition rapidly became worse, with greater cyanosis, deep quick respirations, and very rapid, scarcely audible heart sounds. The patient died at 9:35 a. m.

Blood cultures made July 4 showed a growth of an anaerobic hemolytic short-chained streptococcus, probably *Streptococcus viridans.*

Necropsy.—Two hours' post mortem.—Body: Emaciated, 165 cm. long. None but hypostatic discolorations of the skin.

Thoracic Cavity: When the thorax is opened the heart is found lying in a pericardial sac distended with fluid. The apex is in the fifth space, 9 cm. to the left of the midsternal line, and the right margin of the ventricle is 6 cm. to the right of the midsternal line. The plurae are smooth except for a small, easily broken adhesion at the right base posteriorly, and contain no free fluid.

Lungs: Left, weight 365 gm. At the apex is a dry, firm, red nodule 7 cm. in diameter. The remainder of the lung is crepitant and mottled with dark, brick-red areas on a yellowish ground. Right, weight 445 gm. The lung appears similar to the left one. Microscopically edema is found, with an infarct, and some areas of atelectasis.

Heart: Weight 252 gm. Right auricle: No dilatation or hypertrophy. The foramen ovale is closed. Some non-adherent, apparently post mortem, blood clot in the appendage. The coronary sinuses normal. Tricuspid valve: 10.5 cm., leaflets thin and covered with smooth endocardium; not retracted; attached to normal chordae tendinae. Right ventricle: Slight dilatation and hypertrophy; the walls measure 5 mm. Pulmonary valve: 7.5 cm. There are only two cusps, which are of equal length, and show no line of former fusion. No thickening or evidence of endocarditis can be seen. The valve apparently could function. A 5 mm. orifice, placed 2.5 cm. above the origin of the cusps, leads by a tube 8 mm. long into the aorta. This open ductus Botalli has just within its lumen a vegetation of fresh appearance, and a collection of similar pinkish vegetations from 1 to 3 mm. high form a streak leading from the orifice over the surface of the pulmonary artery in a posterior and slightly downward direction. The streak is from 1 to 3 cm. wide and aside from the vegetative roughenings the endocardium over it appears corrugated and darkened, but not finely irregular. The orifice of the ductus has no mound-like elevation about it as was described in Wells' case (Case 6 in the tabular summary). The duct is almost cylindrical in shape, but there is slight narrowing of the lumen near the middle. About the aortic mouth and throughout the lumen there is yellowish discoloration but no definite raised patches or sclerosis. On the aortic side the duct opens 5 cm. above the origin of the aortic valves, just beyond the left subclavian, by an orifice 5 mm. in diameter. The opening is smooth except for the presence, just below it, of a straight, ridge-like formation 5 mm. long and 3 mm. high, too stiff and thick to bend and act like a valve. Probably this represents the valve-like fold supposed by Strassman[5] to close the duct when blood pressures change at birth. A few vegetations are about this orifice of the duct. Left auricle: Dilated, but not definitely hypertrophied. The auricular appendage free from clot. Mitral valve: 9.5 cm.; there is slight but definite fibrous thickening of the edges of the cusps, but the endocardium is free from vegetations or ulcerations. Left ventricle: Slight dilatation. The walls are 1.2 mm. thick, and the muscle appears normal. Aortic valve: 8 cm. Two cusps only are visible; one is 4.7 cm. and the other 3.3 cm. The leaflets show evidence of severe acute vegetative endocarditis; there are along the line of closure of the largest cusp four soft nodular masses 1 to 3 mm. in diameter, with punctate elevated pink areas about them. The edge of the leaflet has attached to it two granular leaf-like appendages from 1 to 2 mm. thick and 1.1 cm. long, soft in consistency. Back of the vegetations are areas of fibrous thickening. The other cusp has similar but less well marked lesions. Near the posterior attach-

5. Strassmann, P.: Ueber den Mechanismus des Verschlusses des Ductus Arteriosus (Botalli), Arch. f. Physiol., 1893, p. 566; Anatomische und physiologische Untersuchungen über den Blut Kreislauf beim Neugeborenen., Arch. f. Gynaek, 1893, xlv, 393.

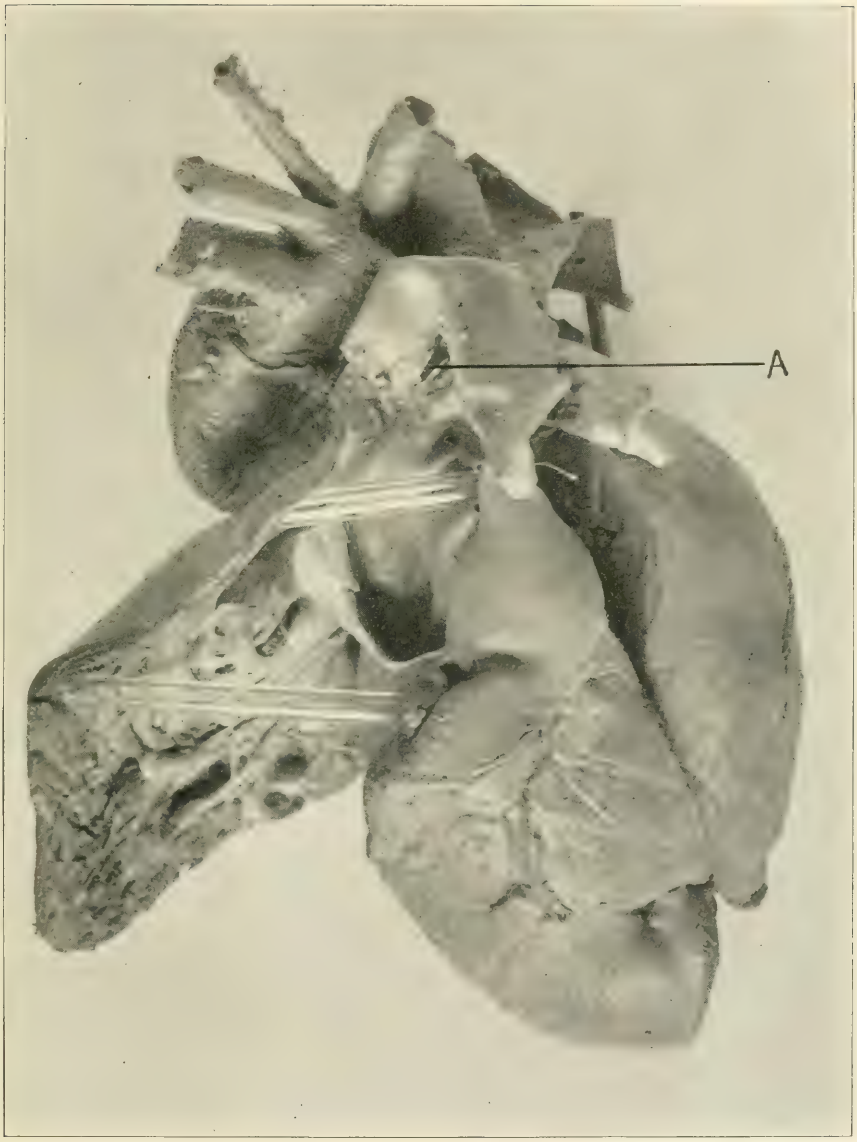


Fig. 2.—View of the heart from the right and above, showing the probe in in the open ductus (*A*), the vegetations on the pulmonary artery to the left of the probe, the four aortic arch branches, and the two cusps of the pulmonary valve.

ment of the anterior cusp is a small ulcerated area 3 mm. in diameter. The sinuses of Valsalva are shallower than normal, due to the retraction of the valves. The right coronary artery opens into the aorta by four small orifices. The arteries are otherwise normal. Aorta: There are several spots of yellowish discoloration but not of definite elevation or increased density. The aorta has four principal branches, the positions and sizes of which are shown in Figure 2. Circumference of aorta 1 cm. above the valve (in formaldehyd hardened specimen) is 6 cm. Circumference of pulmonary artery in corresponding position 6.8 cm. (There is considerable shrinkage in the formaldehyd specimen, but the comparative size of the arteries should be nearly the same, and these measurements show that the pulmonary artery is larger than normal in proportion to the aorta.)

Anatomical Diagnoses.—Open ductus arteriosus (Botalli). Anomalies of pulmonary and aortic valves, consisting in the presence of only two segments in each valve. Four primary aortic arch branches. Acute vegetative, and chronic endocarditis, with connective tissue proliferation, of the aortic valve. Vegetations of the ductus Botalli and pulmonary artery. Slight hypertrophy of the right ventricle. Infarcts of both lungs.

Microscopic examination shows an acute endocarditis of the aortic valves, and endarteritis of the aorta, ductus Botalli and pulmonary artery, with vegetations and masses of streptococci.

A section of the ductus stained with Verhoeff's elastic tissue stain demonstrates a large amount of elastic tissue, thus suggesting that deficiency in elastic tissue is not the only factor in producing patency.

COMMENT

This is one of the rare cases of patent ductus Botalli with "typical" signs, and one of the few cases diagnosed during life and confirmed by necropsy. In the series in the accompanying table only one other of the cases of adults had the lesion included in the antemortem diagnosis.

The endocarditis of the aortic valve above seems to indicate that the vegetations about the ductus Botalli and along the pulmonary artery, and also the lung emboli, owe their ultimate origin to this lesion, and that in fact the infection has made a vivid diagram by its lesions of a course of blood from the aorta through the ductus Botalli.[6] While the embolus which produced the infarction of the lung may have had its immediate origin in the vegetations on the pulmonary artery, it is also possible that it came from the lesions of the aortic valve through the open ductus. The possibility of this form of paradoxical embolism has not received the attention it deserves. It may be nearly as important as the usually cited passages through the foramen ovale, for in 412 cases of congenital cardiac disease patent ductus arteriosus occurred (counting cases with other congenital lesions) 106 times, as compared with 134 cases of open foramen ovale.

6. A very similar case of acute aortic endocarditis with extension through the duct is reported by Schlagenhaufer: Ztschr. f. Heilk., 1901, xxii, 19.

That the lesion had no great disturbance of function as its sequence is plainly seen by the absence of cardiac symptoms in the case history. Clubbed fingers with slight cyanosis of the nails constitute the only recognized effect. In the heart itself there is slight right ventricular hypertrophy, and on consideration it seems probable that there was a distinct disturbance in the pressure in the pulmonary circuit, the cause for which is of course to be sought in the aortic-pulmonary short circuit, and the result to be seen in the dilated pulmonary artery, and hypertrophied right ventricle, and proved clinically by the palpable and loud pulmonic second sound, which was a striking feature of the heart examination. Very probably the clubbing of the fingers is to be traced to this changed pressure condition.

Were the thrill and murmurs due to the patent ductus Botalli or to the aortic lesion? No fact is conclusive proof one way or the other, but there is an accumulation of probabilities, all of which point in one direction, and which give a basis for a theory very reasonable in appearance. In the first place, why should there be a murmur from the passage of blood through this tube any more than through other branches of the aorta? Of course there is a possibility for the production of a murmur in the interference of the ductus current with the current in the pulmonary artery. Examination of the specimen, however, gives a plainer answer; the ductus comes away from the aorta at a peculiar angle, and at the proximal side of the opening is the sharp ridge described above, an arrangement admirably suited to break up the flow and produce the continuous murmur. Assuming that murmurs are transmitted with the blood flow, this murmur would be heard as in the pulmonary artery. The pathological examination indicated that the aortic lesion was a rather recent one, for there was none of the left ventricular hypertrophy and dilatation of a four years' aortic insufficiency. Clinically, also, the absence of signs of aortic insufficiency at the examination four years before death, and the absence at that time of the cardiac history to be expected in the production of the aortic lesions, point in the same direction. The murmur itself does not correspond to the usual character of an aortic insufficiency murmur, and the continuous humming top thrill would be difficult to explain on that basis. Most probably, then, the continuous thrill and the accompanying murmurs were unconnected with the aortic lesion, but were due to the sharp ridge on the aorta near the orifice of the ductus Botalli. The diastolic murmur, greatest over the aortic area, the systolic thrill at the apex (and perhaps part of the other murmurs) coming at the time other signs of aortic disease were recognized, and at the time when the pathological examination and clinical history indicate aortic disease began, suggests that they were probably due to the aortic lesion.

Case	Age	Sex	Cyanosis	Clubbing	Dyspnea	Heart Examination				Other Details
						Right Border	Left Border	Thrill	Murmurs	
I										
1	26	♀	0	0	0?	Incr.	Normal	Felt only once.	"Machinery" murmur through whole cycle, loudest in 3d lt. space.	Dulness in 2 lt. space P_2+.
2	26	♀	0	.....	In crises with palpitation.	.....	.....	.....	Presystolic roll at apex.	2d sound doubled.
3	32	♂	Antemortem.	0	Antemortem	Normal	Normal	.....	Short presystolic over precordia.	.....
4	35	♂	.....	.....	Slight	.....	.....	.....	.....	2d sound doubled.
5	55	♀	Marked	.....	.....	.....	Normal	.....	Faint systolic at apex.	Dulness in 2d space.
6	42	♂	.....	.....	.....	Incr.	Incr.	Suggested at apex.	Loud blowing systolic in 2d rt. space	P_2+. Dulness in 2d lt. space. Severe icterus.
7	17	♀	Slight	Present	0	Normal	Normal	Through cycle over pulmonic area.	Rough systolic running into diastole over pulmonary area.	P_2+. Dulness and pulsation 2d lt. space.
II										
8	6½*	♂	Present	.....	.....	.....	.....	.....	0	.....
9	6*	♀	Attacks	0	.....	Normal	Normal	.....	0	.....
10	11½*	?	Moderate	Present	On exertion.	Incr.	Incr.	0	Systolic at base antemortem.	.....
11	11½*	♀	0	.....	.....	Incr.	Normal	.....	Systolic over pulmonic area.	.....
12	9†	♀	Slight attacks.	.....	.....	Incr.	Normal	0	Systolic loudest in 2d left space.	.....
13	5†	♀	Marked	.....	.....	Incr.	Incr.	0	Systolic loudest 5th space 3 cm. out.	.....
14	3†	♂	Slight	0	.....	Normal	Normal	0	Rough systolic loudest near apex.	P_2+.....
15	7†	♂	Marked	.....	Present	Normal	Normal	.....	.....	.....
16	6½*	♂	General	.....	.....	Normal	Normal	0	Systolic loudest opposite nipple.	Dulness 2d rib to apex.
17	17‡	..	Present	.....	.....	.....	.....	.....	0	.....
18	2†	..	Present	.....	0	Incr.	Normal	.....	.....	P_2+.....
19	2‡	♂	.....	.....	.....	.....	.....	.....	0	.....
20	5‡	♀	Present	Present	.....	Normal	Normal	.....	Sawing systolic, loud at base.	.....
III										
19	....	..	6	.....	6	Usually Incr.	.....	7 — systolic or continuous.	Almost always loud systolic or continuous.	P_2 usualy +
IV										
34	....	..	29%	2.9%	47%	.....	.....	Systolic over pulm. area —29% Continuous —5.9%	Systolic in pulm. area—38%. Continuous in pulm. area—5.9%	P_2+ in 17.2%

* Weeks. † Months. ‡ Days. ♀ Female. ♂ Male.

1. Mead, K. C.: Jour. Am. Med. Assn., 1910, lv., 2205.
2. Doufour, H., and Huber: Bull. et mém. Soc. méd. d. hôp. de Paris, 1911, xxxi, 510.
3, 4 and 5. Motzfeldt, K.: Deutsch, med. Wchnschr., 1913, xxxix, 2037.
6. Wells, H. G.: Am. Jour. Med. Sc., 1908, cxxxvi, 381.
7. This paper.
8. Weber, F. P.: Proc. Roy. Soc., Sect. Study Dis. Child., 1908, p. 193.
9. Cautley, E.: Proc. Roy Soc., Sect. Study Dis. Child., 1908, p. 34.
10. D'Espine, M. H.: Rev. de méd., 1908, xxviii, 941.

		Autopsy					
Ductus	Heart, Wt.,Gm.	Right Ventricle	Left Ventricle	Pulmonary Artery	Valves	Other Organs	Other Defects
Aortic 10 mm. pulm. 4 mm.	395	Dil. and hyper.	Hyper.	dil.	Slight mitral and aortic sten.	.....	None.
4 mm.	...	Hyper.	Hyper.	.....	Mitral sten. and veg.	.....	None.
8 mm.	590	Hyper.>	Hyper.	dil.	Normal	.....	None.
2 mm.	480	Hyper.>	Hyper.	sl., dil.	Normal	.....	None.
Thrombosed.	700	Hyper.	Hyper.	dil.	Normal	.....	None.
Aortic 8 mm. pulm. 2 mm.	300	Slight hyper.	.....	.....	Normal	Liver cirrhosis	None.
5 mm.	252	Slight hyper.	0	sl., dil.	Aortic veget.	Lung infarcts	Two leaflets on aortic and pulmonary valves. Four aortic branches.
5 mm.	...	Slight hyper.	Slight hyper.	.....	Normal	Atelectatic patches	None.
.....	...	Slight hyp. and dil.	Slight	.....	.....	.....	Slight narrowing of aorta, patent foramen ovale and ventricular septum.
6x3 mm.	95	Hyper. and dil.	.....	.....	.....	.....	Aorta rides ventricles, obliterated pulmonary artery, patent ventricular septum.
.....	...	.....	Hyper.	Lumen ¼ in.	.....	Slight emphysema	Aorta rides ventricles, patent ventricular septum, atresia pulmonary artery.
.....	...	.....	.....	sl., dil.	.....	Pneumonia	Absent ventricular septum, slight narrowing aorta, four aortic branches.
"Small"	...	.....	.....	.....	.....	.....	Absent interauricular septum, fused mitral and tricuspid valves, four aortic branches.
"Small"	...	Dil. and hyper.	.....	.....	.....	Bronchopneumonia	Large ventricular septum defect, small open foramen ovale.
"Mod. size"	...	Hyper.	.....	Atresia	Aortic sten.	Emphysema	Atresia pulmonary artery, open foramen ovale.
.....	90	Hyper.	.....	Atresia dil. above	Sl. aortic sten.	Bronchopneumonia	Atresia pulmonary artery, open foramen ovale.
.....	...	.....	.....	Atresia	.....	.....	Atresia pulmonary artery.
2 mm.	...	Hyper.	.....	Oblit.	Normal	Atelectasis	Obliterated pulmonary artery.
.....	...	.....	.....	.....	.....	.....	Aorta and pulmonary artery arise in common.
1 mm.	...	.....	.....	.....	.....	Bronchopneumonia	
.....	...	Usually hyper.	Often hyper.	Often dil.			
.....	...	Hyper. in 32.1%	Hyper. in 32.1%				

11, 12 and 13. Carpenter, G.: Brit. Jour. Child. Dis., 1908, v, 396.
14. Sawyer, J. E. H.: Birmingham Med. Rev., 1909, lxvi, 152.
15. Bach, S.: Arch. f. Kinderh., 1909, l, 31.
16. Bradley, W. N.: New York Med. Jour., 1909, lxxxix, 1302.
17. Vallois, M.: Bull. Soc. Obst. de Paris, 1910, xiii, 107.
18. Edwards, E. P.: Cleveland Med. Jour., 1911, x, 748.
19. Gierke, E.: Charité Ann., 1907, xxxii, 299.
20. Kingsley, C. R.: Johns Hopkins Hosp. Bull., 1911, xxii, 56.

Table 1.—In the first division of the table are the cases since 1908, of patients over 1 year of age—the youngest happens to be 17; in the second division are the cases since 1908 of under 1 year of age; in the third division are the cases collected by Dr. Abbott, and in the fourth those of E. H. Goodman. The case in this paper is tabulated as Case 7. The table summarizes practically all recorded uncomplicated cases of open ductus Botalli, and a few (occurring since 1908) complicated cases. Notes 1 to 20 are the references to the cases in the order given in the table.

An important point in the physical examination is the mention on June 25 of visible pulsation and dulness in the second left interspace, just after the first sound. This corresponds with the strip of dulness, and the Roentgen-ray shadow so much emphasized by several observers and supposed to be made by the dilated pulmonary artery. Pulsation in the suprasternal notch was noted at the same time.

To compare this with previous cases I shall make use of Dr. Abbott's and Dr. Goodman's summaries, and my own of the cases published since.

Besides these may be mentioned the case of Dr. C. H. Dunn,[7] in which an infant with a murmur and slight cyanosis, but no thrill or heart enlargement, was found at necropsy to have an open ductus; and that of Dr. A. Hayashi,[8] in which a baby of 10½ months, with normal heart boundaries, a visible pulsation in the fourth, fifth and sixth spaces, a palpable systolic thrill, and a loud systolic and soft diastolic murmur over the precordia, was proved post mortem to have excessive hypertrophy of the left ventricle, active dilatation of the right ventricle, dilated auricles, and a ductus Botalli of a 5 mm. diameter.

The most interesting single case was reported by Mead. It is Case 1 in the table. Observations covering a period of three years were made. At first slight lateral enlargement in both directions, with a noisy systolic murmur at the base and apex, transmitted to the right of the sternum and almost to the axilla were found. Two years later a strong thrill in the third and fourth right spaces was noted. An examination by Dr. Thayer of Johns Hopkins showed a long machinery murmur over the right ventricle, strikingly loud at the base and the first left interspace, with a late accentuation—almost diastolic—high up. The right border was 5.5 cm. to the right, the left 8.5 cm. No thrill was felt. An impulse was felt in the second and third spaces. A soft systolic and a soft diastolic murmur were heard at the apex. The pulmonic second sound was loud. The diagnosis was made of septum defect or open ductus arteriosus.

In 1909 paralysis of the right vocal chord was diagnosed.

Roentgen-rays showed a bulge on the left of the heart close to the descending aortic arch, considered to be a hypertrophied and dilated

7. Dunn, C. H.: Trans. Am. Pediat. Soc., 1913, xxv, 237.
8. Hayashi, A.: Monatschr. f. Kinderh., 1912, xi, 224.

right auricle. The patient then had irregularities and a loud aortic murmur, and died after several attacks of faintness.

At necropsy open ductus arteriosus was found, with a thickened ring and a fold of membrane about it. The heart had three ruptures of the right ventricle. The right ventricle was dilated and hypertrophied, the myocardium degenerated, the pulmonary artery twice the size of the aorta; there was slight aortic and mitral stenosis.

In the analysis of cases in the accompanying table it seems evident that characteristic symptoms or physical signs are rare. Cyanosis occurred in 70 per cent. of the cases as against the 31 per cent. of Dr. Abbott, and was about as frequent in the uncomplicated cases as in the whole. Clubbing of the fingers was noted in only 15 per cent.; in 25 per cent. of the cases there was dyspnea, as against Dr. Abbott's 31.5 per cent. A constant definite thrill was noted only once; Dr. Abbott finds it in 37 per cent., and many writers (De la Camp[9] and others) speak of it as one of the most useful diagnostic signs. A systolic murmur over the base, extending into diastole, was noted only twice (both of them cases of adults). A systolic murmur, loudest over the base, was present in five other cases (only one of them a case of an adult). Dr. Abbott, however, finds that a peculiar loud murmur is nearly always produced, almost invariably beginning in systole, and localized near the base of the heart. A murmur of some kind was present in 65 per cent. of the cases above.

Goodmann, in his collection of 34 cases with necropsy, found twenty females and eleven males. Cyanosis was found in 29 per cent., dyspnea in 47 per cent., palpitation in 37 per cent., clubbed fingers in 2.9 per cent., pulsation in the second left interspace in 5.9 per cent., a systolic thrill over the pulmonary area in 29.4 per cent., a systolic and diastolic thrill over the same area in 5.9 per cent., a systolic murmur over the area in 38 per cent., and continuous murmur in 5.9 per cent. The pulmonic second sound was accentuated in 17.2 per cent. The left ventricle was hypertrophied as often as the right—32.1 per cent.

In this series, whether analyzed by classifying the cases according to the division into simple and complicated cases, or into infantile and adult, the sexes are as evenly divided as possible. Dr. Wells, in his forty-one cases, found a remarkable preponderance of females (63 per cent.), which in the light of this series now seems probably was a matter of chance.

Evidently none of the symptoms or signs just discussed are to be depended upon for a constant diagnosis.

---

9. De La Camp, Familiäres Vorkommen Angeborener Herzfehler — Zugleich ein Beitrag zur Diagnose der Persistenz des Ductus Arteriosus Botalli, Berl. klin. Wchnschr., 1903, xl, 48.

The pathological finding of hypertrophy with or without dilatation of the right ventricle was noted in all of the adult cases and in seven of the infantile. Dr. Abbott states that it is the usual pathological finding. Dilatation of the pulmonary artery was found in five of the adult cases and one of the infantile, and clinically the dulness in the second interspace near the sternum emphasized as its result was actually observed four times in the adult cases. (Of course in these statistics it must be remembered that in the infantile cases the examination was evidently often far from thorough.) The pulmonic second sound was noted to be exaggerated in three of the adult cases. It is evident, then, that clinical signs depending on an increased pressure in the pulmonary circuit are the most constant signs in this series. They are common to so many other conditions, however, dilatation of the pulmonary artery often occurring in congenital lesions such as defects of the lower part of the interauricular septum, widely patent foramen ovale, defects of the base of the interventricular septum, transposition of the arterial trunks, stenosis of the aorta, and without other defects, that they do not seem to be of great diagnostic importance.

This analysis indicates that in a large number of cases there is nothing in the way of signs or symptoms to make us sure of an open ductus arteriosus, or even to make us suspect it. Hochsinger emphasizes the difficulty of diagnosis in infants, but hardly in adults. Before a conclusion based on such a small number of cases is accepted, however, it should be determined whether it is supported by reason. What should we expect as the clinical result of the lesion? It does not seem to me that a murmur or thrill is by any means a necessary or even likely result from the slight intermixture of currents arising from the presence of an additional aortic branch at an acute angle, emptying by a small opening into the pulmonary artery. (Hochsinger makes this mixing of currents his main explanation of the origin of the murmur, but there are no facts to support this view except the lack of a murmur in certain cases in which such mixing would not be expected. He has too few cases, however, and murmurs are lacking too often, to allow his arguments to convince.) It is interesting to observe that in the three adult cases in which a thrill or murmur at the base of the heart was noted, there was a distinct fold of endocardium about the aortic orifice of the duct (see description of the pathological findings in this and in Mead's case), and in one case a mound-like elevation about the pulmonary orifice (Wells' case). It seems much more reasonable to consider that the murmur or thrill is not the result of the patency alone, but depends on the presence in addition of some endocardial projection, or other roughening or vegetation, such as is recognized in other situations to give rise to

murmurs. This especially, when it is considered that in the cases with necropsy the ductus was open to almost a constant diameter, thus giving each time about the same anatomical reason for a murmur. The clinical findings of absent murmurs in many cases is in perfect accord with this reasoning. The conditions due to increased pressure in the pulmonary circuit could hardly be expected to be very marked when the small size of the ductus is considered (barely over 5 mm.), and there is of course nothing pathognomonic about them when discovered. Combinations of the signs discussed can be effected by combinations of lesions, which are often found in congenital heart disease. No signs are necessarily present when the ductus is open, then, which are sufficiently specific to afford us means of diagnosis. Even the peculiar murmur occasionally produced accessorily may be closely simulated in pulmonary stenosis, or in defects of the ventricular septum. In occasional cases the peculiar humming systolic murmur, loudest over the pulmonary area, with or without a thrill, combined with signs of increased pressure in the pulmonary artery, such as loud and palpable pulmonic second sound, increased dulness in the second left interspace, increased in the middle Roentgen-ray shadow, and suprasternal pulsation, with or without cyanosis, clubbed fingers, and dyspnea, justify the inclusion of "open ductus" in the differential diagnosis. Of course a continuous murmur over the pulmonary area makes a more certain diagnosis possible. The indefinite murmurs, etc., often present, may give rise to confusion with acquired valvular lesions in a way which can easily be seen from the tabulated cases. The present series of cases brings out the rarity of recognizable symptoms, especially of the thrill and characteristic murmur, rather than new symptoms or signs.

Many articles have been published (Arnheim,[10] De La Camp,[9] Miller,[11] Wessler and Bass[12]) in which it is assumed or claimed that the diagnosis can be made frequently, or almost always, by using the Roentgen-ray and careful percussion to determine the dilatation of the pulmonary artery. It is noteworthy, however, that most of these articles are based on cases without necropsy.

In the cases since 1908 there are only two with roentgenoscopy showing a dilated pulmonary artery confirmed by necropsy; no necropsy was made in the three Roentgen-rayed cases of Dr. Abbott. In view of these facts and of the non-characteristic nature of the findings, any emphasis on this method for diagnosis seems unwarranted.

---

10. Arnheim, G.: Persistenz des Ductus Botalli, Berl. klin. Wchnschr., 1903, xl, 616.

11. Miller, R., and Orton, G. H.: A Case of Open Ductus Botalli with X-Ray Examination, Brit. Jour. Child. Dis., 1913, x, 109.

12. Wessler and Bass: Persistent Ductus Botalli and Its Diagnosis by the Orthodiagraph, Am. Jour. Med. Sc., 1913, clxv, 543.

In regard to the prognosis, the table brings out strongly the apparent fact that the patients that die in infancy are those with other congenital heart lesions, while those that live over one year, have an indefinite term of life. It will be noted that none of the adult patients had another important congenital heart lesion, while of the patients that died under 1 year of age only two failed to have one. The pure cases of open ductus, in other words, have little interference with function.

In spite of all that has been written on the subject, conclusions must be tentative until the extraordinarily small number of cases carefully and thoroughly observed is much increased.

SUMMARY AND TENTATIVE CONCLUSIONS

1. The case presented here is one of the rare cases with those signs of open ductus arteriosus usually regarded as typical.

2. It is one of the rare cases diagnosed during life and confirmed by necropsy.

3. The case illustrates the actual course of blood during life through the ductus.

4. The possibility of a practically unrecognized form of paradoxical embolism is shown.

5. A summary and discussion of cases seems to show that:

(*a*) The physical signs formerly regarded as characteristic are more often absent than present, and the possibility of diagnosis must be rare.

(*b*) Most of the signs discussed are really not absolutely characteristic.

(*c*) Combinations of the signs can occur in combinations of other lesions.

(*d*) When there is the rare combination of signs formerly regarded as diagnostic, the presence of the lesion is probable, but not certain.

(*e*) Far too few cases have had roentgenoscopy and necropsy to determine the value of the Roentgen-rays for diagnosis.

(*f*) The Roentgen rays determine only a dilatation of the pulmonary artery, which is present in several other lesions than open ductus, so that the Roentgen ray findings will not make a certain diagnosis possible.

(*g*) The characteristic murmur is probably not the result of the patency of the duct alone, but requires in addition the presence of endocardial folds, vegetations or other roughenings, about the ductus.

(*h*) The only result of the patency is increased pressure in the pulmonary artery (with its secondary effects).

(*i*) The former view of preponderance of cases in females was merely the result of chance.

(*j*) The open ductus alone does not lead to early death, but when complicated with other congenital heart lesions death is usual within one year.

(*k*) Very few cases have been carefully observed and recorded. More cases are needed before conclusions can be definite.

I wish to express my thanks to Dr. Henry A. Christian, chief of the medical service, for his permission to report the case; and to Dr. W. T. Councilman, for valuable suggestions in the treatment.

In addition to the references mentioned in the text, the following may be consulted:

Zinn, W.: Zur Diagnose der Persistenz des Ductus Arteriosis Botalli, Berl. klin. Wchnschr., 1898, xxxv, 433.

THE CLINICAL ACTIONS OF VERATRUM*

RUSSELL J. COLLINS, M.D.
CLEVELAND, O.

Veratrum is obtained chiefly from the rhizome and roots of *Veratrum viride,* an American plant, although the *Veratrum album,* a European plant, has also been used in medicine. Both varieties contain a mixture of alkaloids, the main action being due to protoveratrin. In the past, veratrum has been employed mainly as a cardiac depressant and to soften the pulse and lower the blood pressure in eclampsia.

According to Wood[1] the effects of therapeutic doses on the circulation of mammals (dog and rabbit) consist of a slowing of the pulse and a moderate fall in blood pressure, the effects being rather persistent. The slowing is due mainly to stimulation of the vagus center (abolished by cutting the vagi). The vasomotor center is not stimulated except by the fall in blood pressure and by respiratory embarassment.

Toxic doses produce at first exaggeration of the vagus stimulation as a marked slowing, irregularity and final arrest, with corresponding fall in blood pressure. This is followed by sudden extreme acceleration and rise of blood pressure (partly asphyxial and partly spasmodic). This rise may last for several minutes, and is succeeded by a rapid progressive fall and death. Other signs of "toxicity" are profuse sweating, nausea, followed quickly by vomiting, diarrhea, dysphagia, collapse, paralysis and light convulsions.

The object of the present study was to ascertain more definitely the effects produced by veratrum in normal and diseased human individuals, with special reference to the circulatory system. Many of the studies reported in the literature lack definite objective data, and whatever data exist need the confirmation of the improved and more modern methods of observation. Then, too, veratrum being a very active drug pharmacologically, there is reason to believe that certain hitherto overlooked therapeutic applications could be made of it. A report of the results thus far obtained is here presented.

---

* Submitted for publication March 12, 1915.

* From the Pharmacological Laboratory, Medical School, Western Reserve University, and the Medical Services of the Lakeside and City Hospitals.

1. Wood, H. C., Jr.: Jour. Am. Med. Assn., 1906, xlvii, 2061.

METHODS

The patients were selected from the wards of Lakeside and City hospitals. All were convalescent with the exception of Patients 4 and 8. During the observations all patients were lying in bed while the pulse rate and blood pressure were taken. The noon day meal consisted of a soft diet.

The pulse was taken for half a minute at the time of the first dose of veratrum and at intervals of fifteen to thirty minutes until the effects of the drug were pronounced. The blood pressure was taken by the auscultatory method before the administration of the drug and again when the pulse rate had reached a minimum. The patient (Case 6) was allowed to walk around for five minutes when the pulse reached 60 with no effect on the rate.

The preparation used was the 10 per cent. tincture from *Veratrum album*.[2] Each dose was given in seven cases with from one to three glasses of water. By mistake one patient (Case 4) received only an equivalent of one-fourth glass and complained that each dose caused considerable gastric irritation, of which none of the others complained. All patients noted a fulness and throbbing in the head when the pulse rate reached its minimum.

PROTOCOLS OF CASES

The following abbreviated protocols contain the essential data obtained with eight patients.

CASE 1.—Convalescent from acute nephritis. Pulse rate for ten days previously ranged from 80 to 100 per minute. Blood pressure on three preceding days at the same time each day was 135 mm. systolic; 95 mm. diastolic. Total dosage of *veratrum album* was 60 minims in three hours. Pulse rate at administration of first dose was 102; systolic blood pressure, 135 mm.; diastolic, 95. One hour following the last dose pulse rate was 63; systolic blood pressure, 92 mm. and diastolic, 60 mm. Thirty minutes later the patient became nauseated and vomited.

CASE 2.—Convalescent from typhoid fever; received a dosage of 20 minims in one hour with no appreciable effect on the pulse rate and blood pressure.

CASE 3.—Convalescent from acute nephritis. Pulse rate for ten preceding days was 66 to 90. Systolic blood pressure had varied from 180 mm. to 190; diastolic, from 125 mm. to 135 mm. At time of first dose pulse rate was 87; systolic blood pressure, 190 mm., and diastolic, 135 mm. Total dose of veratrum was 60 minims. Fifty minutes following the last dose pulse rate was 63; systolic blood pressure, 135 mm., and diastolic, 95 mm. Twenty minutes later patient became nauseated and vomited.

CASE 4.—Hypertonus. As this was the patient's first day in the hospital, no former data could be obtained. Pulse rate at time of first dose was 88; systolic blood pressure, 228 mm. and diastolic, 142 mm. One hour and fifteen minutes after the last dose, pulse rate was 63; systolic blood pressure, 160 mm. and diastolic, 130 mm. Thirty minutes later patient became nauseated and vomited.

---

2. I wish to express my thanks to Prof. Henry Kraemer of the Philadelphia College of Pharmacy, who furnished us with several preparations of different species of veratrum.

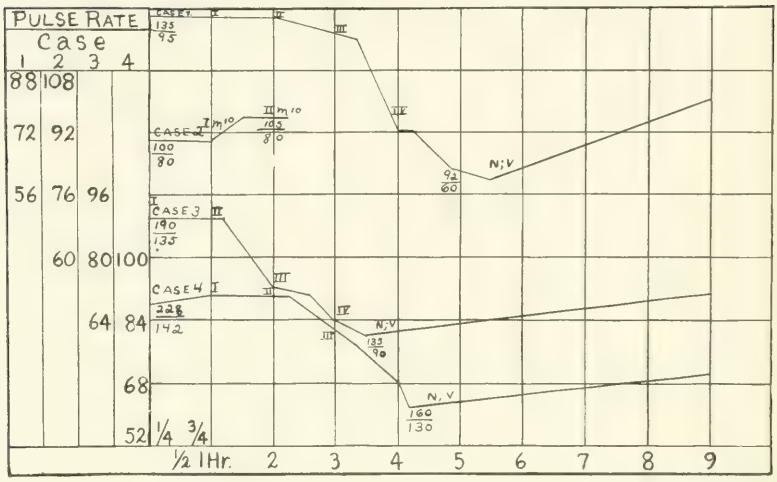


Fig. 1.—Effect of veratrum on pulse rate and blood pressure (individual cases). In the fractions the numerator refers to systolic blood pressure, the denominator to diastolic blood pressure. "H" means headache; "N" nausea; "V" vomiting. The number of the dose is indicated by Roman numerals. Each dose represents 15 minims of tincture of veratrum, except as indicated by Arabic numerals. These data apply also to Chart 2.

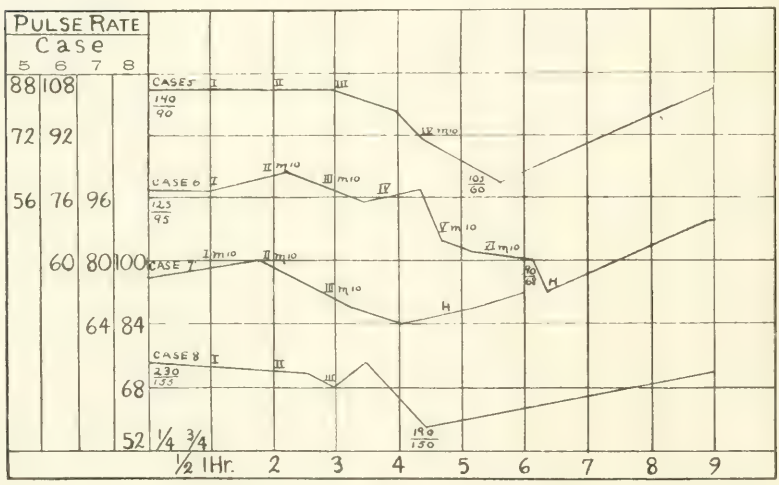


Fig. 2.—Effect of veratrum on pulse rate and blood pressure (individual cases). See legend Chart 1 for additional data.

Case 5.—Convalescent from typhoid fever. Pulse rate on five days previous was 72 to 100. Pulse rate at the time of the first dose was 84; systolic blood pressure 140 mm. and diastolic, 90 mm. Total dose of veratrum was 55 minims in three hours and twenty minutes. One hour and fifteen minutes after the last dose, pulse rate was 60; systolic blood pressure 105 mm. and diastolic 60 mm.; no toxic symptoms.

Case 6.—Convalescent from acute nephritis. Pulse rate for ten preceding days ranged from 80 to 90. Pulse rate at the time of the first dose was 78; systolic blood pressure 125 mm. and diastolic 95 mm. Received 75 minims of veratrum in four hours and five minutes. Pulse rate fifty-eight minutes following the last dose was 52; systolic blood pressure 90 mm. and diastolic 68 mm. The toxic symptoms were only slight nausea and headache.

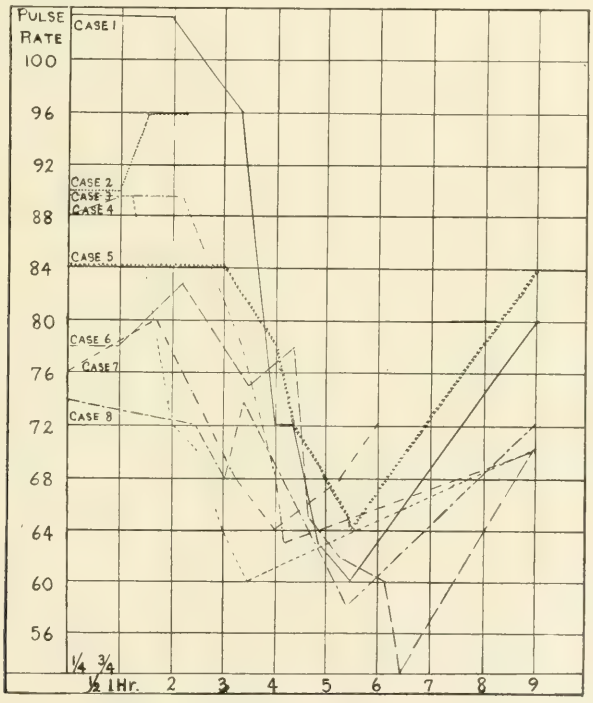


Fig. 3.—Effect of veratrum on pulse rate (composite curve from all cases.)

Case 7.—Tuberculosis of lung; no previous data obtained. At the beginning of observation pulse rate 76. Pulse rate forty-five minutes following the last dose was 64.

Case 8.—Hypertonus and syphilitic aortitis. Pulse rate for ten days previous was 68 to 96; blood pressure had been taken almost daily for a week previously, and ranged from 265 mm. to 230 mm., systolic; 175 mm. to 150 mm., diastolic. Nitrites in the form of nitroglycerin reduced the systolic blood pressure from 265 mm. to 230 mm.; the diastolic from 175 mm. to 150 mm. Total dose of veratrum given was 45 minims during a period of two hours. One-half hour after the last dose, pulse rate was 58; systolic blood pressure 190 mm. and diastolic 150 mm.; no toxic symptoms.

The data from these cases are graphically presented in Figures 1 and 2, and in the form of a composite curve in Figure 3. These indicate that in those patients receiving enough of the drug, that is, from 30 to 75 minims of the tincture, an average fall in pulse rate of 26.9 beats resulted. In all these cases the rate was lowered independent of the original rate. Since three cases of acute nephritis and one case of typhoid were convalescent, and there were, in addition, two cases of hypertonus, a division into two groups will be made. In Group 1, the convalescent cases, the average fall of systolic blood pressure was 39.5 mm.; of the diastolic, 31.75 mm. In Group 2, hypertonus cases, the average systolic fall was 49 mm.; diastolic 8.5 mm. Three cases presented "toxic" symptoms consisting of nausea and vomiting, but in each of these cases the fall in pulse rate and blood pressure preceded the "toxic" symptoms. It has as yet been impossible to determine definitely the duration of action of veratrum in the doses given, although the hospital charts indicate a recovery of from ten to fifteen beats per minute in six hours following the last dose.

SUMMARY

1. The therapeutically effective dose of the tincture of *Veratrum album* for adults ranges from 30 to 75 minims.

2. Clinically, the effects of veratrum resemble the pharmacologic effects, and consist of a slowing of the pulse rate amounting to 12 to 42 beats per minute, and a fall of systolic blood pressure amounting to about 39; of the diastolic, 32 mm. The two hypertonus cases showed a fall of systolic blood pressure amounting to about 49 mm.; of the diastolic about 8 mm.

3. The circulatory effects produced by veratrum take place independently of the "toxic" symptoms, such as nausea and vomiting.

I wish to express my thanks to Prof. Sollmann and Dr. Hanzlik for numerous suggestions and guidance in the work and criticism of the manuscript; to Professors Hoover and Carter for permission to use the material of their clinics at Lakeside and City Hospitals, respectively.

THE RELATION OF CALCIUM TO THE DELAYED COAGULATION OF BLOOD IN OBSTRUCTIVE JAUNDICE*

ROGER I. LEE, M.D. AND BETH VINCENT, M.D.
BOSTON

It has long been recognized that in certain cases of obstructive jaundice there is a tendency to hemorrhage due to the delayed coagulation of the blood. Morawitz and Bierich[1] studied the causes of bleeding in cholemia and came to the conclusion that the altered coagulation time in these cases could be traced to delayed formation of fibrin ferment. They attributed this delay to a diminution or absence of thrombokinase. They believed that the altered coagulation time was not due to the presence of cholates or biliary acids in the blood and was independent of the duration or intensity of the jaundice. Kunika[2] investigated the clinical value of the determination of the coagulation time in cases of icterus and concluded that the delay was due to a decrease in the liver function. King and Stewart,[3] in the course of an investigation of the cause of bradycardia and low blood pressure in dogs with obstructive jaundice, found that the calcium content of the blood was increased and suggested that in icteric conditions the calcium might be combined with bilirubin and biliverdin as a protective mechanism against the toxic effect of these bile pigments. King, Bigelow and Pearce[4] found that the calcium output was increased in obstructive jaundice in dogs. They found an increase in the calcium content of the blood, liver and kidneys. The excess of calcium which appeared in the stools was accompanied by a decrease in the calcium of the other tissues. They did not attempt to study the coagulation in these cases but made the suggestion that the retarded coagulation time may be due to the fact that the calcium is bound to the biliary pigments in such a way as not to be promptly available for the process of clotting.

In a previous article[5] we have studied the processes of coagulation in normal human blood. We have now applied the methods of investigation followed in those studies to the examination of the blood in various types of delayed coagulation, among them the retarded coagulation associated with obstructive jaundice. Our experience coincides

*Submitted for publication, March 9, 1915.

1. Morawitz and Bierich: Arch f. exper. Path. u. Pharmakol., 1907, lvi, 115.
2. Kunika, S.: Deutsch. Ztschr. f. Chir., 1912, cxviii, 574.
3. King and Stewart: Jour. Exper. Med., 1909, xi, 673.
4. King, Bigelow and Pearce: Jour. Exper. Med., 1911, xiv, 159.
5. Lee, Roger I., and Vincent, Beth: The Coagulation of Normal Human Blood, THE ARCHIVES INT. MED., 1914, xiii, 398.

with that of other observers that the coagulation time is not markedly altered in the group of cases called catarrhal jaundice. In this paper we shall limit the discussion to obstructive jaundice without apparent disturbance of liver function. Those cases of liver disease with non-obstructive jaundice present a marked interference with the coagulation of the blood. But according to our present views the reasons for a delay in coagulation in such cases is not the same as in obstructive jaundice with a functioning liver.

The data presented are both clinical and experimental.

CLINICAL OBSERVATIONS

The clinical data were obtained from cases of obstructive jaundice observed in the wards of the Massachusetts General Hospital. A considerable group of cases was studied rather incompletely. Five cases were studied more in detail. It was our experience that there was little or no change in the coagulation time of the blood when taken by the method described by Lee and White,[6] until after the jaundice had been noticed by the patients for periods averaging five weeks. Case 3 of the series will be given in some detail as it is fairly typical of the rest and presents the necessary points for discussion.

Case 3.—Hospital No. 196,455. A man, aged 53, came under observation July 8, 1914. Family and past history, negative. For four weeks the patient has noticed increasing jaundice and weakness, and has lost 12 pounds. There are no other symptoms.

On examination, considerable loss of weight is noted, the skin and sclera are deeply jaundiced; there are small petechial spots on the legs. Chest negative. Liver dulness extends from the fifth rib to 4 cm. below the costal margin where an indistinct edge is felt. Below this edge is felt a rounded mass, taken to be the gallbladder. Examination otherwise negative. Urine negative except for bile. Wassermann negative. The stool was the typical soapy stool with increased fatty acids, but no bile by Hammarsten's test. Gastric examination negative by Ewald test meal and bismuth roentgenogram. On July 10, the coagulation time was seven and a half minutes, which was interpreted as being within the upper limits of normal for that method. On July 18, coagulation time was ten and a half minutes, which was interpreted as being abnormal. For four days he was given calcium lactate 60 grains a day and bilisalol 9 grains a day. On July 24, the coagulation time was eight minutes. Without treatment the coagulation time of the blood became retarded, until July 31, when the time was fifteen minutes. At this time some blood was taken in oxalate and studied according to the methods described in our previous paper.

As a result of this examination it was apparent that cytozyme, serozyme and fibrinogen were present in the patient's blood and active to a normal degree. These points will be discussed more in detail later. There was evidence of a lack of available calcium. This lack of available calcium was then studied in two ways:

1. By contrasting the coagulation time of 1 c.c. of blood taken in the usual way with the coagulation time of the same amount of blood placed in a similar small glass tube to which had previously been added a certain amount

6. Lee and White: Am. Jour. Med. Sc., 1913, cxlv, 495.

of calcium. The amount of calcium which we used arbitrarily was 3 drops of a 1 per cent. solution of calcium chlorid. Control determinations, on normal bloods and on bloods showing a delayed coagulation time due to other causes than obstructive jaundice, showed that the introduction of this amount of calcium made no difference in the controls. In the obstructive jaundice cases, however, there was usually a striking change.

2. The therapeutic administration of calcium, and the subsequent determination of the coagulation time.

In Case 3 on August 1, 1 c.c. of blood clotted by the usual method in thirteen minutes, but in the tube with 3 drops of the 1 per cent. calcium chlorid it clotted in five minutes, which is within the normal limits. On August 1 the administration of 100 grains of calcium lactate per day was begun by mouth. As was our experience in the other cases in this series it was several days before there was any marked change in the coagulation time of the blood.

On August 6 the coagulation time was ten minutes under the usual conditions and five minutes in the tube with the calcium. On August 9, however, the coagulation time was seven minutes under the usual conditions and five minutes in the tube with the calcium. The patient was then considered to be in a condition which warranted operation as regards the danger from hemorrhage.

At operation on August 10 there was no abnormal bleeding although there was an extensive resection of a mass in the common duct which microscopic examination showed to be adenocarcinoma. The patient died of shock and pneumonia in forty-eight hours with no evidence of hemorrhage. There was no necropsy.

The striking facts from the clinical data of this series were the following:

1. The relatively long time after the onset of jaundice before delay in coagulation, averaging five weeks. This probably depends not only on the intensity of the jaundice but also on the amount of calcium in the other tissues which is gradually taken into the blood in the course of five weeks, as King, Bigelow and Pearce have shown.

2. The striking parallelism between the determinations of the coagulation time of the blood and the tendency to hemorrhage especially as seen at the time of operation.

3. The value of the simple control determination of the coagulation time by the use of a second determination with 3 drops of calcium chlorid 1 per cent. to 1 c.c. of blood. This test has been constant and seems to determine the indication for the use of calcium therapeutically. We have called the procedure the "calcium in vitro" test.

4. The apparent necessity for the administration of large doses of calcium by mouth over a period of several days before the coagulation time shows any marked change. This conclusion is based on many observations and many attempts to vary the coagulation time.

LABORATORY OBSERVATIONS

Laboratory experiments were carried out with the blood of the patients in this series. By means of a syringe 18 c.c. of blood were taken from the arm vein directly into 2 c.c. of 1 per cent. oxalate.

Normal blood was taken in a similar fashion as a control. These bloods were put through various tests described in our previous paper. The blood platelets were isolated and found to act normally in the pathological blood, both in hastening the clotting of blood, the formation of thrombin and the retraction of the clot. Serozyme was obtained and found to be efficient in the production of thrombin. The test used for fibrinogen was merely the observation of the firmness of the clot. There seemed to be no difference between the clots of the pathological plasmas and those of the normal controls. The only discrepancy found was in the optimum amount of calcium required to recalcify the oxalated plasma. A typical protocol follows:

The oxalated blood of the patient in Case 3 and a control normal blood were centrifuged at low speed for ten minutes. The control (so-called "cloudy plasma"), on being recalcified with 4 volumes of the recalcifying fluid, clotted in nine minutes. The plasma from the patient in Case 3 clotted in twenty-two minutes. An increase in the amount of calcium in the recalcifying fluid did not appreciably change the clotting time of the cloudy plasma in the normal case but it markedly shortened the clotting time of the pathological blood. The plasma in Case 3 plus 4 parts of the recalcifying fluid clotted in twenty-two minutes. With the addition of 1 drop of 1 per cent. calcium chlorid solution it clotted in seven minutes; 1 c.c. of normal plasma plus 4 parts of the recalcifying fluid clotted in nine minutes. By the addition of 1 to 3 drops of 1 per cent. calcium chlorid solution it clotted in seven minutes.

To cause clotting in some of our cases of obstructive jaundice with a delayed coagulation time, required seven volumes of the recalcifying fluid instead of the normal, which is four. (Four volumes of the recalcifying solution contain one and one-half times the calcium needed to neutralize the oxalate in one volume of the plasma solution, oxalated 1:1,000.)

In order to confirm our laboratory findings, bile was added directly to normal plasma. This bile was obtained from two sources: one, the ordinary powdered ox bile of commerce, taken up with 16 parts of salt solution, the other, sterile bile obtained from a human gallbladder at the time of operation. This bile was diluted 1:4 with salt solution.

The following protocol is typical of many experiments.

Normal "cloudy plasma" plus 4 volumes of the recalcifying fluid clots in sixteen minutes. Four drops of normal plasma, plus 1 drop of bile, plus 4 or 5 volumes of the recalcifying fluid does not clot. Four drops of normal plasma, plus 1 drop of bile, plus 6 volumes of the recalcifying fluid, clots in seventy minutes. If the calcium be gradually increased, clotting can be obtained in twenty minutes. Now if 2 drops of bile be added it requires double the amount of calcium to obtain even feeble clotting. By the addition of an excess of calcium, clotting can be obtained in twenty-four minutes. On the other hand, the clotting time of the recalcified oxalated plasma without bile is not hastened by an increase in the amount of calcium and is delayed when the calcium is in great excess. Four drops of oxalated plasma plus 16 drops of the recalcifying fluid, clots in seventeen minutes; plus 2 drops of 1 per cent. calcium chlorid, in sixteen minutes; plus 4 drops, in twenty-two minutes; plus 10 drops of calcium

chlorid, in thirty-two minutes. If an excess of bile be added there is no clotting no matter how much calcium is added. If the unit be taken as 4 drops of the oxalated plasma, which normally clots on the addition of 4 volumes of the recalcifying fluid in about sixteen minutes, the addition of 3 drops of ox bile in normal salt solution 1:16, or diluted human bile, 1:4, is sufficient to prevent clotting despite the presence of any amount of calcium.

Furthermore, it was found that the effect of weak solutions of bile added after the formation of thrombin could apparently be neutralized by the calcium; but when the bile was present in a considerable amount, an active thrombin would not clot a fibrinogen solution no matter how much calcium was used.

We also attempted to determine whether the action of the bile could be neutralized by cytozyme or serozyme but no results were obtained. An excess of calcium seemed to counteract the effect of bile in weak solutions. On the other hand, in strong solutions, the addition of calcium did not counteract the effect of bile. The action in vitro of a solution of ox bile in normal salt solution was very similar to that of human bile obtained from a gallbladder. It seems probable that the amount of bile in the blood corresponds closely to the mixture of plasma with a weak solution of bile.

EXPERIMENTAL OBSTRUCTIVE JAUNDICE

A condition of obstructive jaundice was produced experimentally in a dog. Repeated observations were made on the coagulation time; when the coagulation time showed definite retardation the therapeutic effect of calcium both by mouth and intravenously was tested. The details of the experiment were the following:

A female dog weighing 5,000 gm., was operated on under ether anesthesia on Sept. 13, 1914. The common bile duct was doubly ligated and 0.5 cm. of the duct resected between the ligatures. A complete obstructive jaundice resulted and persisted until the end of the experiment. At the end of twenty-four hours the urine showed bile, after forty-eight hours the stools became light colored and repeated examination showed the absence of bile by Hammarsten's test. The coagulation time was determined before operation. One c.c. of blood plus 3 drops of sodium chlorid solution clotted in three minutes; 1 c.c. of blood plus 3 drops of 1 per cent. calcium chlorid clotted in three minutes.

On September 27, two weeks after operation, the coagulation time of blood taken from the jugular vein was as follows: 1 c.c. of blood, plus 3 drops of sodium chlorid solution, clotted in six and a half minutes; 1 c.c. of blood, plus 3 drops of 1 per cent. calcium chlorid solution, clotted in six and a half minutes.

On October 23, six weeks after operation, the dog had lost considerable weight but was lively and seemed well. Weight 4,200 gm. Coagulation time—jugular vein—1 c.c. of blood, plus 3 drops sodium chlorid solution, clotted in nine minutes; 1 c.c. of blood, plus 3 drops of 1 per cent. calcium chlorid solution, clotted in four minutes.

October 25, coagulation time—jugular vein—1 c.c. of blood, plus 3 drops of sodium chlorid solution, clotted in twelve minutes; 1 c.c. of blood, plus 1 drop of 1 per cent. calcium chlorid solution, clotted in twelve minutes; 1 c.c. of blood, plus 3 drops of 1 per cent. calcium chlorid solution, clotted in four minutes;

1 c.c. of blood, plus 5 drops of 1 per cent. calcium chlorid solution, clotted in three and a half minutes. At 1:30 p. m. the dog was given, intravenously, 20 c.c. of calcium lactate, 2 per cent., in sterile salt solution. There was no apparent effect. One hour later, coagulation time, jugular vein: 1 c.c. of blood, plus 3 drops sodium chlorid solution, clotted in four minutes; 1 c.c. of blood, plus 1 drop of 1 per cent. calcium chlorid solution, clotted in seven minutes; 1 c.c. of blood, plus 3 drops of 1 per cent. calcium chlorid solution, clotted in seven minutes; 1 c.c. of blood, plus 5 drops of 1 per cent. calcium chlorid solution, clotted in seven minutes.

On October 26, coagulation time—jugular vein—was as follows: 1 c.c. of blood, plus 3 drops of sodium chlorid solution, clotted in six minutes; 1 c.c. of blood, plus 3 drops of 1 per cent. calcium chlorid solution, clotted in five minutes.

On October 27, the coagulation time—jugular vein: 1 c.c. of blood, plus 3 drops of sodium chlorid solution, clotted in eight and a half minutes; 1 c.c. of blood, plus 3 drops of 1 per cent. calcium chlorid solution, clotted in five minutes. At 12:30 p. m. the animal was given, intravenously, 15 c.c. of a 2 per cent. calcium lactate solution in normal salt. The coagulation time—jugular vein—one hour later, was as follows: 1 c.c. of blood, plus 3 drops of sodium chlorid solution, clotted in seven minutes; 1 c.c. of blood plus 3 drops of calcium chlorid, 1 per cent., clotted in five minutes.

On October 28, coagulation time of blood: 1 c.c. of blood, plus 3 drops of sodium chlorid solution, clotted in twelve minutes; 1 c.c. of blood, plus three drops of 1 per cent. calcium chlorid, clotted in seven minutes.

On October 29, coagulation time—jugular vein: 1 c.c. of blood, plus 3 drops of sodium chlorid solution, clotted in eleven minutes; 1 c.c. of blood, plus 3 drops of 1 per cent. calcium chlorid solution, clotted in five minutes. At 10 a. m., 35 c.c. of a 2 per cent. calcium lactate solution in normal salt was given intravenously. The coagulation time—jugular vein—one and a half hours later, was: 1 c.c. of blood, plus 3 drops of sodium chlorid solution, clotted in nine minutes; 1 c.c. of blood plus 3 drops of 1 per cent. calcium chlorid solution, clotted in five and a half minutes.

The dog was then given calcium lactate by mouth in capsules to the amount of 50 grains a day for three days. At the end of that time, the coagulation time—jugular vein—was as follows: 1 c.c. of blood, plus 3 drops of sodium chlorid solution, clotted in six and a half minutes; 1 c.c. of blood, plus 3 drops of 1 per cent. calcium chlorid solution, clotted in seven minutes. The next day the animal was found dead in the cage. Necropsy at 2 p. m., in brief, showed a thin dog with bile-stained tissues. There was no evidence of hemorrhage; the peritoneal cavity contained 250 c.c. of cloudy fluid. The peritoneum was lightly injected with occasional flakes of fibrin. Over the liver was considerable shaggy fibrin. Numerous small abscesses were found in the liver. The common bile duct was found permanently interrupted. On opening the proximal end of the duct purulent material and bile were found. The heart cavities were filled with a firm post-mortem clot. The organs were macroscopically negative except for the staining with bile. Microscopical examination was negative. Culture from the heart showed a few colonies of staphylococcus albus; culture from the liver abscesses showed a scum growth of staphylococcus albus.

It is perhaps unfortunate that the picture was complicated by the low grade of sepsis, but we doubt if this sepsis affected the results in the slightest degree.

In general the results in this experiment correspond closely with our clinical experience. In both conditions obstructive jaundice resulted in a delay in the coagulation time of the blood. This delay, even when the obstruction was complete, was not apparent immediately but gradually increased with the persistence of the jaundice and seemed

to reach its maximum only after about five weeks. It was evident that, experimentally, clinically, and in vitro, this delay in coagulation could be counteracted to a large extent by calcium. In the dog and in human beings the administration of calcium by mouth in sufficient quantity seemed effective. In the dog calcium given intravenously had a very prompt but rather temporary effect on the coagulation.

SUMMARY AND CONCLUSIONS

Obstructive jaundice in the presence of a liver functioning in an adequate manner causes a delay in the coagulation time of the blood. This condition must be differentiated from certain somewhat similar conditions in which the liver itself is seriously damaged. The delay in the coagulation of the blood in obstructive jaundice is apparently due to a lack of available calcium in the blood and can be counteracted by the administration by mouth of calcium salts. Other workers have demonstrated an increase of calcium in the blood in obstructive jaundice and have suggested the plausible explanation that the bile pigments in the blood form a more or less loose union with the calcium salts and so render the calcium unavailable for immediate use in the process of coagulation. Our clinical experience shows that the need of more calcium can be very simply demonstrated at the bedside by the "calcium in vitro" test which we have described.

The results of our studies would tend to show further that while the effect of bile in delaying coagulation is largely counteracted by calcium, yet bile has in addition an inhibitory effect on the formation of thrombin and on the action of thrombin already formed. Moreover, bile in very strong concentration in vitro entirely prevents coagulation even in the presence of an excess of calcium. It is to be doubted, however, if in obstructive jaundice such concentration of bile ever occurs as will entirely prevent coagulation of blood.

Clinically this need of calcium can be met by the administration of any calcium salt. We have mainly used calcium lactate in the dosage of 100 grains a day. We must administer calcium over a period of several days before any marked effect on the coagulation time is seen. It is evident that the necessity of employing large doses depends on the difficulty of securing the absorption of calcium from the gastrointestinal tract rather than the need of such large amounts of calcium.

In a dog, and presumably in human beings, prompt effect on the coagulation time can be obtained by the intravenous injection of calcium salts in solution. Apparently calcium can be given in this way without bad results, but its effect on the coagulation of the blood is transitory.

From a clinical point of view it would seem desirable in all cases of obstructive jaundice to make several determinations of the coagula-

tion time by the methods we have described. In these cases of obstructive jaundice relatively large doses of calcium should be given by mouth in order to meet the excessive demand for calcium which is present in this condition. The early administration of large doses of calcium seems particularly important in cases in which a deficiency in the available calcium as shown by the "calcium in vitro" test and in cases which may subsequently demand surgical interference. When an immediate effect is desired the intravenous administration of calcium is indicated and seems to have no bad effects.

COMPLEMENT FIXATION IN PERTUSSIS*

MIRIAM OLMSTEAD, A.M., AND PAUL LUTTINGER, M.D.
NEW YORK

Since Bordet and Gengou in 1906[1] announced the discovery of the etiological cause of whooping cough, basing their claim mainly on the complement fixation reactions given by the serum of whooping cough convalescents, complement fixation has been studied by many serologists both from an etiological and from a diagnostic standpoint.

Bordet and Gengou used as antigen a saline emulsion of a culture of the Bordet-Gengou bacillus on solid medium. Four hours at room temperature were allowed for fixation, and readings were made the following day. In later publications[2] the constancy of a positive complement fixation reaction among convalescents is reaffirmed, but no figures are given. The work of Bordet and Gengou has been confirmed more or less completely by some investigators and by others their claim has been declared invalid. Meier[3] found the serum of pertussis patients to react with an extract of the lung tissue of patients dying of pertussis. Arnheim[4] using an antigen of the Bordet-Gengou bacillus, obtained positive reactions in six out of twelve cases and thought imperfect technic might account for the lack of a positive reaction in the other cases. In a later publication[5] he stated that these twelve cases were tested again and also three others, and that by making the readings immediately instead of on the following day, as before, he obtained a positive reaction in twelve of the fifteen. The antigen consisted of an equeous emulsion heated at 58 to 60 C. for one hour, shaken on the following day for three hours and centrifuged. Fraenkel[6] tested the serum of five pertussis convalescents against an antigen of the Bordet-Gengou bacillus and obtained a positive reaction

---

* From the Research Laboratory, Health Department.

* Submitted for publication Feb. 9, 1915.

1. Bordet and Gengou: Le microbe de la coqueluche, Ann. de l'Inst. Pasteur, 1906, xx, 731.

2. Bordet and Gengou: Note complémentaire, Ann de l'Inst. Pasteur, 1907, xxi, 720; Note Complémentaire sur le microbe de la coqueluche et sa variabilité au point de vue du serodiagnostic et de la toxicité, Centralbl. f. Bakteriol., orig., 1912, lxvi, 276.

3. Meier, G.: Anwendung der Komplementbindungsmethode bei Keuchhusten, Deutsch. Med. Wchnschr., 1907, xxxiii, 1558.

4. Arnheim, G.: Ueber den gegenwartigen Stand der Keuchhusten Frage, Berl. klin. Wchnschr., 1908, xlv, 1453.

5. Arnheim, G.: Keuchhustenuntersuchungen, Arch. f. Kinderh., 1909, 1, 296.

6. Fraenkel, C.: Untersuchungen zur Entstehung des Keuchhustens, München, med. Wchnschr., 1908, lv, 1683.

in only one, though his technic was apparently the same as Bordet's. Seiffert[7] mentions a positive complement fixation reaction given by one case of whooping cough, against an antigen of the Bordet-Gengou bacillus. Klimenko[8] has reported one case, convalescent three weeks, that gave a positive reaction with a Bordet-Gengou antigen. Wollstein[9] tested the serum from nine patients, in the first to the ninth week of the disease, against antigens of the Bordet-Gengou bacillus and the influenza bacillus, and all results were negative. She used as antigens salt solution suspensions of the Bordet-Gengou bacillus prepared according to the method of Bordet and Gengou, extracts of the bacilli, made by suspending the growth of three blood-agar slants in 5 c.c. of salt solution and shaking twenty-four hours in the thermostat, and extracts from the lungs of a case that came to necropsy, prepared according to the method of Wassermann and Meier. Menschikoff[10] reported positive results with the serum of two pertussis convalescents, using antigens of the Bordet-Gengou bacillus. Later Baecher and Menschikoff[11] found the serums of whooping cough cases examined in the first to the sixth week of the paroxysmal stage, to have no complement fixation power; but positive reactions were obtained with the serums of cases repeatedly vaccinated. Four hours at room temperature were considered necessary for the fixation of complement. The antigen used was a saline emulsion of Bordet-Gengou bacilli. Twenty-one treated cases were tested, nine of which gave a positive reaction. Finizio[12] tested the serum of eight convalescents and obtained six positive reactions with an antigen of the Bordet-Gengou bacillus. Poleff[13] tested two cases in a late stage of whooping cough, using as antigen a saline suspension of two strains of the Bordet-Gengou bacillus; and the results were negative. Imai[14] in testing the blood of convalescents by complement fixation confirmed the results of Bordet and Gengou.

---

7. Seiffert, G.: Ueber den Bordetschen Keuchhustenbazillus, München. med. Wchnschr., 1909, lvi, 131.

8. Klimenko, W. N.: Die Aetiologie des Keuchhustens, Centralbl. f. Bakteriol., orig., 1909, xlviii, 64.

9. Wollstein, M.: The Bordet-Gengou Bacillus of Pertussis, Jour. Exper. Med., 1909, xi, 41.

10. Menschikoff, W.: Ueber den Erreger des Keuchhustens, Russk. Vrach., 1909, p. 1044.

11. Baecher and Menschikoff: Ueber die Aetiologische Bedeutung des Bordetschen Keuchhustenbacillus und den Versuch einer spezifischen Therapie, Centralbl. f. Bakteriol., orig., 1911, lxi, 218.

12. Finizio, G.: Der Bordet-Gengousche Bacillus in der Aetiologie des Keuchhusten, Ztschr. f. Kinderh., orig., 1911, iii, 121.

13. Poleff, L.: Ueber den Bordet-Gengouschen Keuchhustenbacillus, Centralbl. f. Bakteriol., orig., 1913, lxix, 23.

14. Shiga, Imai and Eguchi: Eine Modification von Bordet-Gengous Nahrboden für die Keuchhustenbacillen nebst einigen Ergebnissen in serologischer Beziehung, Centralbl. f. Bakteriol., orig., 1913 lxix, 104.

Bordet and Gengou[15] in 1911 called attention to the value of complement fixation in diagnosing cases of pertussis without a whoop or other typical symptom and mentioned one such case, an adult. Gengou and Brunard[16] had previously reported positive complement fixation reactions with the serum of three adults who had been coughing for two months and who undoubtedly had whooping cough. Delcourt[17] had likewise demonstrated the diagnostic value of complement fixation by obtaining positive reactions in seven doubtful cases of pertussis. Netter and Weil,[18] working with the Bordet-Gengou bacillus, found complement fixing substances to develop too late to be of service in the diagnosis of whooping cough in normal cases. With one exception (a child that had been whooping six days) they obtained no positive reactions even by the use of active serum earlier than the second week of the whoop. During the second week active serum was found to give more positive reactions than inactive. After the fifteenth day all sixteen cases tested reacted positively, whether active or inactive serum was used. In regard to the duration of the complement fixation reaction, Weil[19] claimed that a positive reaction was always given within three months of cure and might be given thirteen years after cure. He based this statement on a study of nine patients who had been cured for from two months to thirteen years. Weil suggests that other diseases, for example, measles, may have an effect on the complement fixation reaction in pertussis. Of three cases of measles that had been whooping three to five weeks, he found two to give a negative complement fixation reaction with a Bordet-Gengou antigen and the other one to give only a weakly positive reaction. Friedlander and Wagner[20] have reported positive complement fixation results in all stages of pertussis, including the early catarrhal, and consider the test of great diagnostic value. Their antigen is a saline emulsion of a seventy-two hour growth of the Bordet-Gengou bacillus on ascitic fluid agar. They have used active serum. A water bath at 37 C. was used for fixation and incubation and readings were made within an hour after the addition of cells and amboceptor. Friedlander and Wagner

15. Bordet and Gengou: Le diagnostic de la coqueluche fruste par la methode de la fixation d'alexine, Centralbl. f. Bakteriol., orig., 1911, lviii, 573.

16. Gengou and Brunard: Apropos du diagnostic de la coqueluche chez l'adulte, Bull. de l'Acad. Roy. de méd. de Belgique, 1910, xxiv, 329.

17. Delcourt: Diagnostic de la coqueluche fruste par la Réaction de Bordet-Gengou, Arch. de méd. d. enfants, 1911, xiv, 30.

18. Netter and Weil: La déviation du complément par le bacille de Bordet-Gengou dans la coqueluche, Comp. rend. Soc. de biol., 1913, lxxiv, 236.

19. Weil, M.: La déviation du complément vis-à-vis du bacille de Bordet-Gengou dans la coqueluche, Compt. rend. Soc. de biol., 1913, lxxiv, 260.

20. Friedlander and Wagner: Diagnosis of Whooping Cough by the Complement-Deviation Test, Jour. Am. Med. Assn., 1914, lxii, 1008; Am. Jour. Dis. Child., 1914, viii, 134.

TABLE 1.—Strains Used for Antigens

Strain	History	Characteristics		
		Morphological	Cultural	Serological
P. D.	From another laboratory, originally from Prof. Bordet....	Typical Bordet-Gengou...	Typical Bordet-Gengou...	Fixation with all other Bordet-Gengou strains
55	From sputum of a case in the sixth day of the paroxysmal stage	Typical Bordet-Gengou...	Typical Bordet-Gengou...	Fixation with all other Bordet-Gengou strains
81	From sputum of a very severe case in the fifth day of the paroxysmal stage	Typical Bordet-Gengou...	Typical Bordet-Gengou...	Fixation with all other Bordet-Gengou strains
93	From a throat swab of a case in the eighth day of the paroxysmal stage	Typical Bordet-Gengou...	Typical Bordet-Gengou...	Fixation with all other Bordet-Gengou strains
95	From sputum of a very severe case in the first day of the paroxysmal stage	Typical Bordet-Gengou...	Typical Bordet-Gengou...	Fixation with all other Bordet-Gengou strains
98	From sputum of a case not whooping but developing whoop later	Typical Bordet-Gengou...	Typical Bordet-Gengou...	Fixation with all other Bordet-Gengou strains
100	From sputum of a case in the third day of the paroxysmal stage	Typical Bordet-Gengou...	Typical Bordet-Gengou...	Fixation with all other Bordet-Gengou strains
114	From sputum of a case with doubtful whoop.....	Typical Bordet-Gengou...	Typical Bordet-Gengou...	Fixation with all other Bordet-Gengou strains
121	From sputum of a case in the first day of the paroxysmal stage	Typical Bordet-Gengou...	Typical Bordet-Gengou...	Fixation with all other Bordet-Gengou strains
141	From sputum of a case in the second week of the paroxysmal stage	Typical Bordet-Gengou...	Typical Bordet-Gengou...	Fixation with all other Bordet-Gengou strains
154	From sputum of a case in the second week of the paroxysmal stage	Typical Bordet-Gengou...	Typical Bordet-Gengou...	Fixation with all other Bordet-Gengou strains
155	From sputum of a case in the third week of the paroxysmal stage	Typical Bordet-Gengou...	Typical Bordet-Gengou...	Fixation with all other Bordet-Gengou strains
C.	Probably from pertussis, isolated about two years.....	Atypical Bordet-Gengou..	Atypical Bordet-Gengou. Grows without blood	No cross fixation
1.	From sputum isolated about one year; no record of source	Atypical Bordet-Gengou..	Atypical Bordet-Gengou. Grows without blood	Some cross fixation with Bordet-Gengou strains
31	From sputum of case in third week of paroxysmal stage....	Atypical Bordet-Gengou..	Atypical Bordet-Gengou. Grows without blood	Some cross fixation with Bordet-Gengou strains and with 10
10	From sputum of case in third week of paroxysmal stage....	Atypical Bordet-Gengou..	Atypical Bordet-Gengou. Grows without blood	Some cross fixation with 31
33	From sputum of case in second week of paroxysmal stage	Typical influenza bacillus	Hemoglobinophilic bacillus	Fixation with some other influenza strains
35	From sputum of case in fourth week of paroxysmal stage	Typical influenza bacillus	Hemoglobinophilic bacillus	Fixation with some other influenza strains
37	From sputum of case in second week of paroxysmal stage	Typical influenza bacillus	Hemoglobinophilic bacillus	Fixation with some other influenza strains
87	From sputum of case in first week of paroxysmal stage....	Typical influenza bacillus	Grew several generations on ascitic, then became hemoglobinophilic	Fixation with some other influenza strains
747	From spinal fluid of case of cerebrospinal meningitis.....	Typical influenza bacillus	Hemoglobinophilic bacillus	Fixation with some other influenza strains
B12	From another laboratory.....	Typical influenza bacillus	Hemoglobinophilic bacillus	Fixation with some other influenza strains
Z	From spinal fluid of case of cerebrospinal meningitis.....	Typical influenza bacillus	Probably hemoglobinophilic bacillus	Fixation with some other influenza strains

obtained a positive complement fixation test in all eighteen cases tested in the paroxysmal stage. Of twelve cases in the catarrhal stage, from one to five weeks before the appearance of the whoop, eleven gave positive reactions. One case of doubtful pertussis gave a positive reaction. Sixteen normal individuals were negative.

The Bordet-Gengou bacillus is not the only one that has been found to give a positive complement fixation reaction with the serum of pertussis cases. Manicatide,[21] who claims to have discovered in *Bacillus Z* the etiological factor in pertussis, has found the serum of nineteen whooping cough cases to give a positive complement fixation reaction with an antigen of *Bacillus Z*. Negative reactions were given by six normals.

The variation in the results of different workers may be explained partly by differences in technic, such as, method of preparing antigen, amount of antigen used in test, amount of serum used in test, use of active or inactive serum, time and temperature allowed for fixation, interval between the addition of sensitized cells and the reading of results, method of reading reactions, partly by a difference in the interval between taking and testing the blood specimens, and partly by the small number of cases tested. An explanation of some discrepancies is still to be found. It is difficult to reconcile Manicatide's findings with those of Bordet and his followers.

The objects of our investigation were to test the validity of the claim for the etiological relationship of the Bordet-Gengou bacillus to whooping cough and to determine the diagnostic value of complement fixation in this disease. Various strains of the Bordet-Gengou bacillus, of atypical Bordet-Gengou bacilli and of hemoglobinophilic bacilli have been classified according to their complement fixation reactions by elaborate cross titrations of immune rabbit serum and this work is soon to be described in another article.

The strains[22] used for antigens in the tests with human serum are described in Table 1. The typical Bordet-Gengou bacilli may, according to their reactions with immune rabbit serum, be considered practically identical, closely related to the hemoglobinophilic bacilli but distinct from them. The atypical Bordet-Gengou bacilli are (with the exception of C) more or less closely related to each other and to the typical Bordet-Gengou bacillus. The hemoglobinophilic strains are more or less closely related to each other.

21. Manicatide: Der Komplementbindungsvorgang bei Keuchhusten, Ztschr. f. Kinderh., 1913, vii, 226.

22. Most of the strains were isolated by Dr. A. W. Williams and Miss M. A. Wilson and the morphological and cultural identification was made by them. (Williams, A. W.: The Etiology of Pertussis, Arch. Pediat., 1914, xxxi, 567.)

TABLE 2.—Complement Fixation Reactions of Forty-Eight Inactive Specimens from Forty-Six Unvaccinated Cases of Pertussis

Serial Number	Stage of Disease	Complications	Antigens Used																							
			P. D.	55	81	93	95	98	100	114	121	141	154	155	C	I	31	10	33	35	37	87	747	BI$_2$	Z
7a	Whooping 36 days		+++												—										
7b	Whooping 59 days		+++												—	+	—			++				—	
14a	Whooping 102 days		±												+++	—	—							+	
14b	Whooping 117 days		+++												+	++	+			+++				++	
16	Whooping 66 days		—												+	—	—								
17	Whooping 108 days	Congenital idiocy	—												—	—	—								
18	Whooping 102 days		—												—	—	—								
26	Whooping **84 days**		±												—	—	—			—					
27	Whooping 35 days		—												—	—	—		—	—					
28	Whooping 90 days		±													—	—							+	
30	Whooping 49 days		±												—	—	—		—	—					
31	Whooping 60 days		±												—	—	—		—	—					
32	Whooping 21 days		++																						
33	Whooping 24 days		++																						
34	Whooping 14 days		++																						
35	Whooping 60 days		±						++++																
39	Convalescent 5 mos.		±	±	—	—	—	—	—													+++	—		
40	Convalescent 3 mos.,7 days		+	+		—	±	+	—													±			
41	Convalescent 3 mos.			—		—			—													+			
42	Convalescent 4 mos.,12 days		+	±	—	—			—													+++			
54	Convalescent 2 mos.		-																						
55	Convalescent 4 mos.,6 days		-	±		—																			
56	Convalescent 104 days		—	±		—																			

57	Convalescent 112 days	.....	—	—	.....	—	—	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	—	.....	.....
58	Convalescent 104 days	.....	—	—	—	—	—	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	+	.....	.....	.....
59	Convalescent 143 days	.....	±	+	—	—	—	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	+++	.....	.....	.....
60*	Convalescent 4 mos.	.....	—	—	.....	—	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
62	Convalescent 5 mos.	.....	+++	+++	++	+	±	+++	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	+	—	.....	.....
63	Convalescent 4 mos.	.....	±	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
76	Whooping 4 days	Measles.....	—	.....	.....	.....	.....	—	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	—
77	Whooping 15 days	Measles.....	—	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	±	.....	—	.....	.....	.....	.....	.....	.....	.....	±
78	Whooping 2 days	Measles.....	—	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	±	.....	±	.....	.....	.....	.....	.....	.....	.....	±
79	Whooping 11 days	Measles.....	—	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	—	.....	.....	.....	.....	.....	.....	.....	.....	.....	—
80	Whooping 46 days	Scarlet fever	±	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	±	.....	—	.....	.....	.....	.....	.....	.....	.....	—
81	Whooping 11 days	Scarlet fever	—	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	±	.....	±	.....	.....	.....	.....	.....	.....	.....	—
82†	Whooping 3 days	.....	—	.....	.....	.....	.....	.....	.....	—	.....	.....	.....	.....	.....	.....	—	.....	±	.....	.....	.....	.....	.....	.....	.....	—
83	Whooping 2 wks.	.....	.....	.....	±	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	—	—	.....	.....	.....	—	.....	.....	.....	.....
84	Whooping 24 days	.....	.....	.....	+	.....	.....	±	.....	.....	.....	±	.....	.....	.....	±	.....	—	—	.....	.....	.....	.....	.....	.....	.....	.....
95	Whooping 5 wks.	Scarlet fever, chickenpox, otitis media	.....	.....	+++	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	±	.....	.....	.....	.....	.....	.....	.....	.....
96	Whooping 30 days	Broncho-pneumonia	.....	.....	+++	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	+	.....	.....	.....	.....	.....	—	.....	.....
97	Whooping 3 wks.	.....	.....	.....	+	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	—	.....	.....	.....	.....	.....	.....	.....	.....
98	Whooping 14 days	.....	.....	.....	+++	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	—	.....	.....	.....	.....	.....	-	.....	.....
99	Whooping 4 wks.	Tubercular glands	.....	.....	+	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	±	.....	.....	.....	.....	.....	-	.....	.....
100	Whooping 5 wks.	.....	.....	.....	+	++	.....	+	.....	.....	++	.....	.....	±	—	—	.....	—	.....	—	.....	±	—	.....	.....	.....	.....
101	Convalescent 5 days	.....	.....	.....	+	++	.....	+	.....	.....	++	.....	.....	+	—	—	.....	—	.....	±	.....	±	—	.....	.....	.....	.....
104	Whooping 5 days	.....	.....	+++	+++	+++	.....	+++	.....	.....	++++	.....	.....	+++	—	—	+	—	.....	.....	—	—	.....	.....	.....	.....	.....
105	Whooping 2 mos.	Diphtheria; otitis media	.....	+	—	±	.....	—	.....	.....	.....	.....	—	.....	—	—	—	.....	.....	.....	.....	±	.....	.....	.....	.....	.....
106	Whooping 23 days	.....	.....	+++	±	+	.....	±	.....	.....	.....	+++	.....	—	±	-	.....	.....	.....	.....	.....	—	.....	.....	.....	.....	.....
108	Convalescent 2 wks.	Diphtheria...	.....	.....	.....	.....	+	+	.....	.....	.....	.....	.....	+	.....	±	.....	.....	.....	.....	.....	±	.....	.....	.....	.....	.....
109	Convalescent 1 wk.	Pneumonia..	.....	.....	.....	.....	--	—	.....	.....	.....	.....	—	—	.....	—	.....	.....	.....	.....	.....	±	.....	.....	.....	.....	.....

* Strain 55 was obtained from this case.

† Strain 141 was obtained from this case.

Antigens have been prepared by Dr. O. Povitzky as follows: For the four atypical Bordet-Gengou strains, which grow on plain veal agar, Dr. McNeil's method of preparing gonococcus antigen has been successfully employed. Twenty-four hour cultures on salt free veal agar are washed off with sterile neutral distilled water, the emulsion is heated in a water bath at 56 C. for three hours, and at 80 C. for one hour and filtered through a Berkefeld filter. Influenza antigens are prepared from a twenty-four to forty-eight hour growth on 1 to 500 blood veal agar, washed off with sterile neutral distilled water, shaken for two to four hours, left in a thermostat at 56 C. for about eighteen hours and centrifuged or filtered through a Berkefeld filter. Antigens are prepared from the typical Bordet-Gengou strains in the same manner except that Bordet-Gengou potato-blood-agar medium is used and the growth, instead of being washed off, is scraped off with a platinum spud and deposited in the distilled water, in order to avoid extraction from the medium of substances that would render the antigen anticomplementary and nonspecific.

Inaba's[23] antigen, prepared by emulsifying three loops of a forty-eight hour culture in 5 c.c. of physiological salt solution, shaking one to two hours for two days and centrifuging, has been tried but has proved to be no more satisfactory than our antigens; that is, no more specific and of no more value in reacting with the serum of pertussis cases. The antigen described by Friedlander and Wagner,[24] a saline emulsion of a seventy-two hour growth on ascitic agar, has also been tried. The tests on whooping cough cases have been too few to be conclusive, but thus far this antigen has given no more positive reactions than ours and has seemed to be less specific. It has given strong + (doubtful) reactions with serums from normal adults, and it has cross-fixed with the serums of rabbits immunized against atypical Bordet-Gengou strains and influenza strains. Furthermore, it has the disadvantages of all live culture antigens: it must be fresh and it must be standardized immediately before use.

Antigens are standardized at the temperature to be used for tests by determining the antigen unit with a homologous immune serum and the anticomplementary dose of the antigen. For tests one-quarter the anticomplementary dose is chosen, provided that amount gives complete inhibition with the homologous serum; and the antigen is diluted so that 0.1 c.c. contains the desired amount. Best results, strongest and most specific, are obtained with an antigen of long range. An antigen of short range has occasionally been used in an amount greater than one-quarter the anticomplementary dose, but the results are likely to be unsatisfactory.

For a hemolytic system we have used sheep cells in a 5 per cent. suspension, rabbit amboceptor and guinea-pig complement in a 10 per cent. dilution. The system is carefully standardized daily by means of an amboceptor titration and for tests between two and three units of amboceptor are employed. The erythrocyte suspension is sensitized before use. The size of the test is one-tenth that of the classical Wassermann, 0.5 c.c. instead of 5 c.c.

23. Inaba, I.: Ueber den Bordet-Gengouschen Keuchhustenbacillus, Ztschr. f. Kinderh., 1912, iv, 252.

24. Friedlander and Wagner: Diagnosis of Whooping Cough by the Complement-Deviation Test., Am. Jour. Dis. Child., 1914, viii, 134.

The patient's serum is tested in two amounts, 0.02 c.c. and 0.01 c.c. and the anticomplementary property of the serum is determined by 0.04 c.c. and 0.02 c.c. controls. All specimens have been tested inactive and a few comparative tests have been made with active serum.

For fixation a water bath or incubator at 37 C. has been used for some tests on all specimens, and the results of these tests only are given in the accompanying tables. A few comparative tests have been made at room temperature for four hours and at ice-box temperature for four hours. For the second incubation, the water bath is invariably used and the tests are read as soon as the controls, double the amount of antigen used with the serum and double the maximum amount of serum used, are completely hemolyzed. Citron's[25] standard of readings is followed and only those tests are considered positive in which 0.02 c.c. of serum gives complete inhibition of hemolysis.

Tests have been made on specimens of serum from one hundred eleven pertussis cases or suspected pertussis cases (Table 5). Nineteen of these specimens were received when no Bordet-Gengou antigens were on hand and so were tested against atypical Bordet-Gengou strains and hemoglobinophilic strains only. With these antigens no serum, inactive, gave a positive reaction. Of eleven specimens tested active, three gave a positive reaction with strain C and one with strain BI_2.

Tables 2, 3 and 4 contain the results of tests by water bath or incubator fixation on the inactivated serum of the ninety-two specimens that were tested against Bordet-Gengou antigens. The specimens are divided into three groups, namely, those from unvaccinated cases of pertussis (Table 2), those from vaccinated cases of pertussis (Table 3), and those from prophylactic cases, i. e., cases that received vaccine before or after exposure to whooping cough and that did not develop a whoop (Table 4). Forty-eight unvaccinated cases were tested, thirty-two in the first to the seventeenth week of the paroxysmal stage and sixteen in the first to the twenty-first week of convalescence. Twelve of the whooping cases gave a positive reaction with at least one Bordet-Gengou antigen, a percentage of 37.5; and two of the convalescents gave a positive reaction, a percentage of 12.5. The earliest case tested had been whooping two days. The earliest case that gave a positive reaction had been whooping five days. The total number of vaccinated cases tested was thirty-two, six in the fourth to the seventh week of the paroxysmal stage and twenty-six in the first to the twelfth week of convalescence. Three of the whooping cases gave a positive reaction with at least one Bordet-Gengou antigen, a percentage of 50, and fifteen of the convalescent cases, a percentage of 57.7. The highest percentage of positives among unvaccinated cases has been obtained in the paroxysmal stage, among vaccinated cases the highest percentage occurred after the cessation of the whoop. The number of cases is insufficient, however, to establish this as a rule.

25. Citron: Immunity. Translated by Garbart., 1914, p. 184.

TABLE 3.—Complement Fixation Reactions of Inactive Serum from Twenty-Nine Vaccinated Cases of Pertussis

Serial Number	Stage of Disease	Complications	Vaccine Administered§	Amount Vaccine in Millions	Interval Between Last Vaccination and Complement Fixation Test	Antigen Used																						
						P. D.	55	81	93	95	98	100	114	121	141	154	155	C	I	31	10	37	87	747	BI_2	Z
13	Whooping 42 days	.....	P. D.	200	1 mo.	--	.....	.....	.....	.....	.....	.....	...	.....	...	...	.....	—	—	—	...	...	.....	...	—	..
29	Whooping 21 days	.....	P. D.	100	10 days	+	.....	.....	.....	.....	.....	.....	...	.....	...	...	.....	...	.....	+	...	...	.....	...	+	.
37	Convalescent 56 days	Chronic bronchitis	1	860	34 days	++++	++++	++	+++	.....	.....	+++	...	.....	...	...	.....	...	.....	...	...	...	±	...	—	.
38	Convalescent 56 days	.....	1	1,860	34 days	—	—	.....	.....	.....	.....	—	...	.....	...	...	.....	...	.....	...	...	...	.....	...	.....	..
43	Convalescent 53 days	.....	1	10,000																						
			2	1,500	48 days	+	++	.....	+	±	.....	±	...	.....	...	...	.....	...	.....	...	...	...	+++	...	—	..
44	Convalescent 83 days	....	1	875	86 days	—	—	—	—	.....	—	.....	...	.....	...	...	.....	...	.....	...	...	...	+	...	—	..
46	Convalescent 83 days	.....	1	875	83 days	±	+	.....	±	—	.....	—	...	.....	...	...	.....	...	.....	...	...	...	.....	...	.....	..
47	Convalescent 55 days	Tonsillitis....	1	2,400	53 days	++++	++++	.....	++++	.....	.....	.....	...	.....	...	...	.....	...	.....	...	...	...	.....	...	.....	..
48	Convalescent 85 days	.....	1	825	86 days	+	+++	.....	—	.....	.....	.....	...	.....	...	...	.....	...	.....	...	...	...	.....	...	.....	..
49*	Convalescent 47 days	.....	1	12,500																						
			2	2,500	39 days	—	±	—	—	—	.....	.....	...	.....	...	...	.....	...	.....	...	...	...	+	...	—	..
50	Convalescent 50 days	.....	1	8,000	50 days	++++	++++	.....	+++	+++	.....	.....	...	.....	...	...	.....	...	.....	...	...	...	.....	...	.....	..
51	Convalescent 52 days	.....	1	4,150	53 days	+++	+++	.....	—	.....	.....	.....	...	.....	...	...	.....	...	.....	...	...	...	.....	...	.....	..
52	Convalescent 36 days	.....	1	66,500																						
			2	11,000	38 days	+++	+++	.....	—	.....	.....	.....	...	.....	...	...	.....	...	.....	...	...	...	.....	...	.....	.
53	Convalescent 31 days	.....	1	750																						
			2	8,500	30 days	+++	+++	.....	±	±	.....	.....	...	.....	...	...	.....	...	.....	...	...	...	.....	...	.....	.
61	Convalescent 57 days	.....	1	4,850	57 days	--	--	.....	.....	—	.....	.....	...	.....	...	...	.....	...	.....	...	...	...	.....	—	.....	..

64†	Convalescent		1	100																					
	20 days		2	18,700	10 days	++++	++++			+++		++++											+	−	
67	Convalescent		1	4,350																					
	36 days		2	4,000	36 days	+++	+++			−													±		
68	Convalescent		1	5,350																					
	47 days		2	2,000	36 days					−													+++	−	
72‡	Convalescent		1	3,850																					
	36 days		2	6,000	31 days	±	±	±		−	±													−	
85	Convalescent 2 wks.		3					+++			±		−		+			−		−	−				
86	Convalescent 1 wk.		3					+++			+		−		+			−		±	±				
87	Convalescent 1 wk.		P. D.					±			+				−					−	−				
88	Convalescent 19 days		1																						
			3					±																	
90	Convalescent 1 wk.		3					+++			+							±		−	−				±
91	Convalescent 1 mo.		5					+			±							−		−	−				+
93	Convalescent 13 days		3					++			±									−	−				−
94	Convalescent 11 wks.		3					+++			±							−							+
102	Whooping 27 days	Broncho pneumonia							+++														−		
103	Whooping 25 days	Broncho pneumonia	6				+++	+++	+++		+++			+++			+	−	−	−	−	−	−		
107	Convalescent 1 wk.		8	250						+	±					−	+		±				±		
110	Whooping 5 wks.	Penumonia	8	1,975						++++	++++					±	++++		++++				−		
111	Whooping 3½ wks.		8	750						+	±						+						−		

* Strain 87 was obtained from this case.
† Strain 100 was obtained from this case.
‡ Strain 98 was obtained from this case.

§ Vaccine 1—Strains P.D., 33, 35, 37. Vaccine 2—Strains P.D., C, I. Vaccine 3—Strains 55, 93, 98, 100. Vaccine 5—Autogenous. Vaccine 6—Strains 55, 93, 95, 98 Vaccine 7—Strains 55, 93, 95, 98; differs from Vaccine 6 in method of preparation. Vaccine 8—Strains 154, 155, 163.

The fact that the convalescent vaccinated cases were tested within the first three months of convalescence and most of the convalescent unvaccinated later, should be taken into consideration in comparing the two series (Table 6).

Two unvaccinated cases (7 and 14) were tested at two different times. The serum of Case 7 was on both occasions positive with the Bordet-Gengou antigen P. D. Case 14 gave a ± reaction the first time, a ++++ reaction the second. This probably does not indicate a development of complement fixing substances between the first and the second bleedings, in the fifth and the seventh week, respectively, of the paroxysmal stage, but is due to the fact that the first specimen was not tested promptly and had lost antibody content when the test was made. This is the only specimen that, inactive, gave a positive reaction with antigen C; forty-seven were tested. The second specimen of Case 14 (14 b) gave a positive reaction with Antigen 1; only one other specimen (110) out of twenty-nine tested against Antigen 1 gave a positive reaction with this strain. Cases 7 b and 14 b were the only specimens out of nine to give a positive reaction with the hemoglobinophilic strain 35. Case 14 b also gave a positive reaction with BI_2, which appears to be less closely related than any other hemoglobinophilic strain studied to the Bordet-Gengou bacillus; tests on thirty-five other specimens from pertussis cases were negative or doubtful. Whether these reactions of 7 and 14 were nonspecific, due to undeveloped technic, or the poor antigens with which the work was begun, or whether they were due to a mixed infection in the cases, is not known. All tests with antigens 31, 10, 33, 37, 747 and Z have been negative or doubtful. The positive reactions given by antigen 87 are of special interest, as this strain was obtained from the sputum of a pertussis case. Of twenty-seven specimens tested against this strain, five (18.5 per cent.) have given positive reactions, and one of these (43) also gave a positive reaction with the Bordet-Gengou strain 55. The total number of specimens tested against antigens of hemoglobinophilic strains is sixty; the total number giving a positive reaction is six.

There is a marked lack of uniformity among the reactions given by different Bordet-Gengou strains with the same serum. This may be partly due to an inherent difference in the strains that has not been apparent in the cross-titrations; this point is to be investigated later. One reason is a difference in the range of the antigens; the longer the range of an antigen, the more likely it is to give the maximum number of specific fixations. Another reason for the lack of uniformity, unfortunately only recently observed, is the unstability of an antigen after the addition of the salt. The aqueous extract antigens that have been used are stable for many months, but after being made isotonic they

TABLE 4.—Complement Fixation Reactions of Inactive Serum From Twelve Vaccinated Cases Which Did Not Develop Into Typical Pertussis

Serial Number	Symptoms	Vaccine Administered	Amount Vaccine in Millions	Interval Between First Vaccination and Complement Fixation Test	Antigens Used												
					P. D.	55	81	93	95	98	100	C	31	10	87	747	BI_2
36	Slight cough	1	1,040	50 days	±	+	.....	±	+	.....	—	.....	.....	.....	+	.....	—
45	Cough	1	850	2 mos.	—	—	.....	.....	—	.....	.....	.....	.....	.....	+	—	—
65	Occasional cough	1	1,750	39 days	—	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
66	None	1	650	57 days	—	—	.....	.....	—	.....	.....	.....	.....	.....	.....	—	.....
69	Cough for two weeks	1 2	4,350 1,000	43 days	+++	++	.....	.....	—	.....	.....	.....	.....	.....	.....	—	.....
70	Bronchitis	1 2	3,350 2,500	36 days	±	—	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
71	None	1	850	57 days	+	—	.....	.....	—	.....	.....	.....	.....	.....	.....	—	.....
73	Cough two weeks	1 2	4,350 2,500	36 days	±	—	.....	.....	—	.....	.....	.....	.....	.....	.....	—	.....
74	Slight cough	1	650	57 days	—	—	.....	.....	—	.....	.....	.....	.....	.....	±	.....	.....
75	Slight cough	1	650	43 days	—	—	.....	.....	—	.....	.....	.....	.....	.....	+	.....	.....
89	Cough	3	.....	.....	.....	.....	±	.....	.....	—	.....	.....	±	—	.....	.....	+
92	Cough	5	.....	.....	.....	.....	+	.....	.....	—	.....	—	—	—	.....	.....	+

lose fixing power and cannot be depended on for more than two or three days. This fact undoubtedly accounts for some of the negative and doubtful reactions among convalescent cases and those late in the paroxysmal stage. Some serologists would probably consider positive many of the + reactions, those in which there is only slight hemolysis in the tube containing 0.02 c.c. of serum. Many of these reactions undoubtedly are specific; but inasmuch as a reaction of the same strength may be given by an antigen of a closely allied organism that is not the infecting agent, it seems wise to hold to the rule of calling only ++, +++ and ++++ reactions positive. Only cases of pertussis have given ++, +++ and ++++ reactions. The presence of natural antisheep amboceptor in the patients' serum has not been taken into consideration and may account for some doubtful and negative reactions.

TABLE 5.—Summary of Complement Fixation Reactions Among 111 Cases of Pertussis Including Twelve Doubtful Cases Designated as Prophylactic

		Number Positive	Percentage Positive
Number cases tested	111		
Cases not tested against Bordet-Gengou antigens	19		
Cases tested against Bordet-Gengou antigens	92	33	35.9
1. Vaccinated cases of pertussis	32	18	56.2
Vaccinated cases whooping	6	3	50.0
Vaccinated cases convalescent	26	15	57.7
2. Not vaccinated cases of pertussis	48	14	29.2
Not vaccinated cases whooping	32	12	37.5
Not vaccinated cases convalescent	16	2	12.5
3. Doubtful cases of pertussis—vaccinated	12	1	8.3

Complicating infections, such as measles and scarlet fever, may, as suggested by Weil,[19] cause a weakening of the complement fixation reaction for whooping cough. We have tested only four cases suffering from both measles and pertussis. The four all gave negative reactions with antigen P. D., but as they had been whooping only two, four, eleven and fifteen days, respectively, this result was not to be wondered at. The three scarlet fever cases had been whooping one, five and six weeks, respectively, and the second gave a positive reaction with antigen 81. Owing to the low percentage of positive reactions among cases of pertussis without complications, these tests prove nothing as to the effect of other infections on the antibodies of the Bordet-Gengou bacillus.

Among sixty-seven specimens of serum from normals or from patients suffering from some disease other than pertussis (chiefly syphilis or gonococcus infection), no positive reaction has been given by inactive serum with a Bordet-Gengou or an atypical Bordet-Gengou antigen. Tests with influenza antigens have also been negative, with the exception of two positive reactions with BI_2 and one with 87. As an influenza infection in these three individuals prior to the taking of the blood specimens could not be excluded, there was no proof of the nonspecificity of the reactions.

TABLE 6.—Complement Fixation Reactions Among Sixteen Unvaccinated and Twenty-Six Vaccinated Convalescent Cases of Pertussis *

Weeks of Convalescence	Not Vaccinated (16 Cases)	Vaccinated (26 Cases)
1	+ —	+ + — —
2	—	+ +
3		+ —
4		+ —
5		+ + —
6		— —
7		+ + +
8	—	+ + — —
11		+
12	—	+ —
13	—	
15	— —	
16	— — —	
17	—	
18	—	
20	+ — —	

* Each + symbol denotes one case having positive reaction. Each — symbol denotes one case not having positive reaction.
Total not vaccinated = positive 2 or 12.5 per cent. + + — — — — — — — — — — — — — —
Total vaccinated = positive 15 or 57.7 per cent. + + + + + + + + + + + + + + + — — — — — — — — — — —

That the administration of vaccine has an effect on the complement fixation reaction of pertussis cases seems indisputable, but it is doubtful if complement fixing substances would develop in the serum as the result of vaccine alone. Among the twelve vaccinated children that did not develop typical whooping cough (Table 4), the serum of one only, who had coughed for two weeks, gave a positive complement fixation reaction with two Bordet-Gengou antigens (a percentage of 8.3) and this was probably a case of whooping cough running a short atypical course. Two normal adults, inoculated with large doses of

TABLE 7.—Results of Successive Increasing Doses of Pertussis Vaccine on Complement Fixation in the Case of Two Normal Adults

	Date of Injection	Amount of Vaccine 1 Injected in Millions	Reaction	Date of Bleeding	Date of Test	Results											
						Antigen P. D.		Antigen 98		Antigen C		Antigen 10		Antigen BI_2		Antigen Z	
						Active Serum	Inactive Serum	Active Serum	Inactive Serum	Active Serum	Inactive Serum	Active Serum	Inactive Serum	Active Serum	Inactive Serum	Active Serum	Inactive Serum
O. (Had pertussis 21 yrs. ago.)				1/ 1/14	4/ 7/14	++++	—	.....	.....	.....	.....	+++	—	+++	—	.....	.....
	4/ 1/14	125															
	4/ 4/14	250	Local.....	4/ 7/14	4/ 7/14	±	—	.....	.....	.....	.....	—	—	—	—	.....	.....
	4/ 7/14	500	Strong local.....														
	4/11/14	1,000	Local and slight general.														
	4/21/14	2,000	Strong general; headache, dizziness, etc.	4/29/14	5/ 1/14	Strong +	+	.....	.....	.....	±	.....	±	.....	.....	Strong +	±
L. (Never had Pertussis.)				1/ 1/14	4/ 7/14	Strong +	—	—	—	.....	.....	—	—	±	—	.....	.....
	4/ 1/14	125															
	4/ 4/14	250	.....	4/ 7/14	4/ 7/14	—	—	—	—	.....	.....	—	—	—	—	.....	.....
	4/ 7/14	500	Local.....														
	4/11/14	1,000	Strong local.....	4/14/14	4/20/14	.....	+	—	—	.....	.....	.....	.....	.....	.....	.....	.....
	4/14/14	2,000	Slight general; temperature 99.1.														
	4/21/14	4,000	Temperature 100.2; muscular weakness.	4/29/14	5/ 4/14	Strong +	+	.....	.....	.....	±	.....	±	.....	.....	+++	±

vaccine (Table 7), at no time gave a positive complement fixation reaction when inactive serum was used. Before the first inoculation a positive reaction was given by the active serum of O., who had had pertussis twenty-one years before, with antigens P. D., 10 and BI_2. The active serum of L., who never had had pertussis, gave a positive reaction after the sixth injection with antigen Z but several months before this experiment was performed his active serum gave a positive reaction with antigen C.

TABLE 8.—Results of Seventy-Seven Comparative Complement Fixation Tests with the Active and Inactive Serum of Thirty Specimens from Cases of Pertussis

Antigens	Number of Specimens Tested	Number of Specimens Giving Positive Reaction	
		Serum Inactive	Serum Active
P. D.	10	0	2
C	22	1	4
I	9	0	0
31	12	0	0
10	1	0	0
87	1	0	0
BI_2	16	0	3
Gonococcus	6	0	5

These tests on the serum of normal adults demonstrate the danger of making false diagnoses by using active serum. In determining the etiology of pertussis, active serum is of little value. We have tested thirty specimens from pertussis cases, active as well as inactive, some with several antigens, so that seventy-seven comparative tests have been made (Table 8). Of ten tests with P. D. two more positives were obtained with active serum. Of twenty-two tests with C. three more positives were obtained with active serum. Of sixteen tests with BI_2, positives were obtained with active serum only, three in number. Five specimens from whooping cough cases with no history of gonococcus infection gave a positive reaction with a specific gonococcus antigen when tested active; the inactivated serum reacted negatively or doubtfully. Most of the specimens of active serum used as negative controls in the pertussis tests gave negative or doubtful reactions with antigens of the Bordet-Gengou bacillus, but even an occasional false positive is sufficient to destroy the diagnostic value of the test. A negative reaction given by active serum is stronger evidence of the lack of a specific infection than is a negative reaction given by inactive serum;

but a positive reaction given by active serum is, in our experience, no proof of the presence of a specific infection.

The tests made at room temperature for four hours and at ice-box temperature for four hours have been too few to warrant the drawing of conclusions concerning the value of either method as compared with water bath or incubator fixation; but they are sufficient to show that not all convalescent cases of whooping cough give a positive reaction even when four hours are allowed for fixation. Eighteen specimens (eight vaccinated convalescents, nine unvaccinated convalescents and one vaccinated doubtful case) were tested against Bordet-Gengou antigens by both water bath and room temperature methods and four more positives (22.2 per cent.) were obtained by the latter method. Two of these were among vaccinated convalescents, one was an untreated convalescent and the fourth was a prophylactic case. Five vaccinated convalescents gave positive reactions by both methods; one vaccinated convalescent was negative by both; eight untreated convalescents were negative or doubtful in both. Other tests were made that could not be read at all, owing to the anticomplementary action of the antigens. When the temperature of the room is so variable that a standardization of the antigen is no guide to the amount of antigen to be used in the test, this method is not of practical value. We have not tested enough negative controls by the room temperature method to be certain of the specificity of the reaction. Experience in the diagnosis of other infectious diseases by means of complement fixation led us to suppose that constant and reliable results would be obtained by ice-box fixation and a higher percentage of positives than by water bath or incubator fixation. The number of tests made is insufficient to establish this. Thirteen specimens have been tested against Bordet-Gengou antigens, allowing four hours for fixation. Only two more positives (15.4 per cent.) were obtained by this method than by water bath fixation. One was in the case of a vaccinated convalescent, the other an untreated convalescent. One specimen gave a positive reaction by both methods. Of the nine specimens that gave a negative or doubtful reaction by both methods one was from a vaccinated whooping case, one from a vaccinated convalescent and the other seven from untreated cases still whooping. A longer period than four hours would probably give equally specific results and more positive reactions.

As our technic improved the percentage of positive reactions given by the serum of pertussis cases has increased, but even among pertussis convalescents the number of positive reactions is still far below 100 per cent. Even though the power of fixing complement with antigens of the Bordet-Gengou bacillus is not constantly present in the

serum of these cases, the frequency of a positive reaction is so great as to be presumptive evidence of the etiological relationship of the Bordet-Gengou bacillus to whooping cough.

SUMMARY

1. The most reliable antigen for complement fixation tests in whooping cough is obtained by autolyzing an aqueous emulsion of a twenty-four to forty-eight hour growth of the Bordet-Gengou bacillus for eighteen to twenty-four hours at 56 C. and shaking for several hours. The closeness of relationship of the Bordet-Gengou strains is still under investigation. To obtain the maximum number of positive reactions it may be necessary to use a polyvalent antigen.

2. Active serum may give non-specific positive reactions. A negative reaction given by active serum is stronger evidence of the lack of an infection than is a negative reaction given by inactive serum.

3. For fixation one-half hour in the water bath or one hour in the incubator is absolutely reliable. Ice-box fixation probably gives reliable results; the optimum period is still to be determined. Fixation at room temperature for four hours is unsatisfactory, at least in rooms with variable temperature.

4. About 40 per cent. of whooping cough cases have given a positive reaction with antigens of the Bordet-Gengou bacillus, when inactive serum was used. The highest percentage of positives is given by convalescent vaccinated cases. Ten per cent. of whooping cough cases have given a positive reaction with antigens of hemoglobinophilic strains.

5. A ++, +++, or ++++ reaction by inactive serum with an antigen of the Bordet-Gengou bacillus is diagnostic of whooping cough, a + or ± reaction is suspicious, a negative reaction has little significance.

CONCLUSIONS

Complement fixation tests on serum from one hundred eleven cases of pertussis or suspected pertussis support the theory that the Bordet-Gengou bacillus is the etiological factor in the disease. The complement fixation test may be of value in the diagnosis of doubtful cases of pertussis.

40 East Sixteenth Street.

THREE CASES SHOWING CHANGES IN THE LOCATION OF THE CARDIAC PACEMAKER ASSOCIATED WITH RESPIRATION *

FRANK N. WILSON, M.D.
ANN ARBOR, MICH.

THE EFFECT OF VAGUS STIMULATION ON THE PACEMAKER

Under experimental conditions, the heart rate in certain animals varies with each phase of respiration and the relationship is such that the longest diastolic pauses occur during expiration, and the shortest pauses during inspiration. Clinically, exactly similar changes in heart rate are common in children and adolescents, and in adults who are nervous or who are recovering from acute illnesses. They are increased by deep breathing and under such conditions are almost universal. The fact that these changes in heart rate disappear after atropin, or after section of the vagi in animals, demonstrates that they are vagal in origin.

Recent experimental studies by Meek and Eyster[1] and by Lewis, Meakins and White[2] have shown that stimulation of the right vagus may cause the pacemaker to migrate from the upper to the lower portion of the sino-auricular node. Stronger stimulation of the right vagus may, in a small percentage of animals, produce an auriculoventricular rhythm.[3,4] After an auriculoventricular rhythm has been produced by this or other means, stimulation of the vagus may cause the pacemaker to shift from the upper portion of the auriculoventricular node near the coronary sinus to some lower portion.[3] Meek and Eyster[1] have explained these phenomena by assuming that the number of fibers supplied by the vagus to the system of specialized tissue found in the heart diminishes as we move from the sinus node downward.

---

* Submitted for publication February 26, 1915.

* From the Department of Internal Medicine, University of Michigan.

1. Meek, W. J., and Eyster, J. A. E.: Experiments on the Origin and Propagation of the Impulse in the Heart. IV. The Effect of Vagal Stimulation and of Cooling Upon the Location of the Pacemaker Within the Sino-Auricular Node, Am. Jour. Physiol., 1914, xxxiv, 368.

2. Lewis, T., Meakins, J., and White, P.: The Excitatory Process in the Dog's Heart. Part I: The Auricles, Phil. Trans. Royal Soc., London, Series B, 1914, ccv, 375.

3. Meek, W. J., and Eyster, J. A. E.: Experiments on the Origin and Propagation of the Impulse in the Heart. III. The Effect of Vagal Stimulation on the Location of the Pacemaker, the Location of the Pacemaker in Auriculoventricular rhythm, and the Effect of Vagal Stimulation on this Rhythm, Heart, 1914, v, 227.

4. Lewis, T.: The Effect of Vagal Stimulation Upon Atrioventricular Rhythm, Heart, 1914, v, 247.

ECTOPIC RHYTHMS

Rhythms which originate at some point in the heart other than the sinus node are called ectopic rhythms. They have been divided by Lewis[5] into two classes.

Ectopic rhythms of the first class are characterized by a sudden onset and a rapid rate. The pacemaker shifts abruptly to a new location, which may or may not lie in the specialized tissue. Lewis, who believes that the ectopic impulses in such cases are elaborated by pathologic processes, has called these rhythms heterogenetic.

Ectopic rhythms of the second class are characterized by a comparatively slow rate. The change in heart rate at the onset of the new rhythm is gradual. The change in the location of the pacemaker may be gradual or abrupt. The point of origin probably always lies within the specialized tissues. These rhythms are believed to arise in the following way. The rates of the inherent rhythms of the various parts of the system of specialized tissue diminish as we go from the sinus node downward. That portion of the specialized tissue which possesses the highest degree of automaticity at any particular time acts as pacemaker for the heart. When the rate of the sinus rhythm is reduced below the inherent rhythm of some lower center, the latter becomes pacemaker. The same thing occurs when impulses sent out by the sinus node fail to reach the lower centers because of conduction changes, as, for instance, in complete heart block, in which case the ventricular part of the a-v conduction system acts as pacemaker for the ventricles. Lewis has called ectopic rhythms of this type homogenetic. He believes that the impulses which give rise to them are elaborated by physiologic processes. While it is often possible to say with certainty that a given ectopic rhythm is heterogenetic, as for instance in auricular flutter, or homogenetic, as in idioventricular rhythm, this distinction is not always possible if the rate at which the ectopic impulses are formed closely approaches that of the normal sinus rhythm.

At the onset of a homogenetic ectopic rhythm, two centers may for a short time send out impulses at very nearly the same rate. These stimuli arising from different parts of the auricle at about the same time, may meet in the auricular walls causing interference phenomena. Furthermore, at the onset of an a-v rhythm the auricles may respond to the sinus node while the ventricles respond to the a-v node. As the rates of the two centers become divergent, however, one center gains the upper hand and becomes the pacemaker of the whole heart.

5. Lewis, T.: Exceptional Types of Slow Heart Action, Quart. Jour. Med., 1912-13, vi, 221.

During the course of an ectopic rhythm of the homogenetic type, gradual changes in the location of the pacemaker within the specialized tissues analogous to those observed experimentally sometimes occur in man. Two cases recently reported by Weil[6] showed a gradual migration of the pacemaker within the a-v node. Possibly this was produced in one of the cases by vagal pressure which was made shortly before the curve was taken. In a third case reported by the same author, the analysis of the curves was difficult, but in addition to the above changes there seemed to be an alternation of sinus and a-v rhythm and possibly also a migration of the pacemaker within the sinoauricular node.

CASE REPORTS

The following cases are unusual in that changes in the location of the pacemaker may be definitely correlated with respiration.

CASE 1.—*History.*—Mrs. E. D., an American housewife, aged 34, came to the University Hospital, Jan. 12, 1915, because of an ulcerative lesion of the nose and was admitted to the dermatological service. The family history was negative. The patient had had the ordinary diseases of childhood, but had been otherwise well, except for an attack of "rheumatism" at the age of 28. Her symptoms at that time were severe pains in the legs below the knees, with swelling of the shins and violent headaches. These symptoms were associated with a general skin eruption. The lesion on the nose for which she came to the hospital appeared as a small papule in November, 1914, and gradually grew larger and ulcerated. The patient had been short of breath on exertion for about seven or eight months, but had had no other cardiac symptoms. The examination showed an undernourished woman with an ulcerative lesion affecting both alae of the nose, and a similar lesion on the left foot.

Examination.—The heart apex was located in the fifth intercostal space, 1.5 cm. outside the midclavicular line. A marked presystolic thrill was felt over this region. Percussion showed that the right border of the heart extended 1 cm. beyond the sternal edge. There was a marked presystolic shock over the entire precordium. On auscultation a loud presystolic murmur was heard at the apex and the first sound was loud and snapping. The pulmonic second sound was accentuated. Extrasystoles occasionally interrupted the otherwise regular rhythm. The clinical diagnosis was tertiary lues and mitral stenosis. The patient was treated with neosalvarsan and during a stay of three weeks in the hospital the nose and foot lesions showed marked improvement. Electrocardiograms were taken on many different occasions.

CASE 2.—*History.*—Mr. C. L., an American bookkeeper of 23, was examined as an out-patient, March 8, 1915, at the request of the Department of Genito-Urinary Surgery, in which department he was being treated for gonorrheal urethritis and rheumatism. He complained of attacks of tachycardia. The family history was negative except that one aunt died of tuberculosis of the lungs. The patient was said to have had a damaged heart valve at birth. He had had measles, scarlet fever, and a questionable typhoid during childhood. He had also had several attacks of quinsy, the first at the age of 12 and the last in the autumn of 1914. Between the ages of 10 and 12 he began to have attacks of tachycardia during which his heart rate reached 170 or more. These

6. Weil, A.: Beitrage zur klinischen Electrokardiographie, Deutsch. Arch. f. klin. Med., 1914, cxvi, 486.

attacks had persisted up to the time of examination. They always began and ended suddenly; exercise seemed to bring them on at first, but had not appeared to do so for the last five years. The attacks usually lasted for five or six hours and during them he felt faint and weak. In 1910 he had an attack nearly every week but since that time they had become less frequent and he had gone as long as three months without an attack. The last three attacks that the patient had had were more severe than any previous ones; the last one lasted three days and confined him to bed. The patient had stopped attacks at various times by quickly changing his position or by holding his breath. In December, 1914, he contracted gonorrhea and one week later, Jan. 3, 1915, he developed a gonorrheal arthritis of the right wrist.

Examination.—The heart apex was located in the fifth intercostal space, 1 cm. outside the midclavicular line. The systolic impulse at this point was well sustained and was accompanied by a short thrill. The right border of the heart was not beyond the sternal margin on percussion. On auscultation, at the apex, there was a slight presystolic roughening and the first sound was loud and snappy. The second sound was accentuated. Over the fourth left costal cartilage a third sound was distinctly heard when the heart rate was slow. This sound occurred in middiastole and sounded like an echo of the second sound. It was transmitted to within 2 cm. of the apex and upward to the third rib. There was a marked respiratory irregularity during forced respiration. The clinical diagnosis was mitral stenosis. Electrocardiograms were taken frequently over a period of six weeks.

Case 3.—*History.*—Mr. L. D., a student of 22, was examined in the outpatient department on account of palpitation and tachycardia. His family history was negative. He had had measles during childhood, but had never had any of the other common contagious diseases. He had had an attack of tonsillitis one month previous to the examination. He denied venereal disease. He had had periods of rapid heart action for as long as he could remember. These attacks usually lasted about one week. They never began or ended suddenly. The most rapid heart rate that he had ever had was during the last attack, which began about one week before the examination. At this time his heart rate reached 140. He complained that his heart skipped beats occasionally.

Examination.—The heart apex was located in the fifth intercostal space, just inside the midclavicular line. There was no enlargement on percussion and no murmurs were heard on auscultation. The first sound was loud and a third sound could be heard in middiastole when the heart rate was slow during the expiratory phase of deep respiration. Electrocardiograms were taken on several occasions.

RESPIRATORY CHANGES IN THE LOCATION OF THE PACEMAKER IN OR NEAR THE SINUS NODE

The first electrocardiograms taken soon after Patient 1 entered the hospital showed no abnormalities aside from occasional auricular extrasystoles, and a very tall broad P complex such as is commonly seen in mitral stenosis. It was noted, however, that a marked slowing of the pulse occurred on deep breathing, and a large number of records were taken during forced respiration. Figures 1, 2, 3 and 4 are examples of the curves obtained at this time.

Shortly after the beginning of expiration the heart rate slowed to less than one-half its usual rate and then gradually returned to normal. During this slowing the P complex became gradually smaller and

bifurcated in the second and third leads. The flattening of P was sometimes so marked in the third lead that it almost disappeared (Fig. 4). This was never observed in the second lead. In the first lead P showed no appreciable change in height, although it became somewhat broader (Fig. 1).

We have observed similar though less marked changes in the P complex in a number of patients with respiratory irregularities and changes equally as marked have been observed both experimentally and clinically by a number of authors.

A decrease in the size of the P complex has been noted in dogs by Einthoven[7] during vagus stimulation (Lead II), by Eyster and Meek[8] after poisonous doses of morphin (Lead II), and by Blumen-

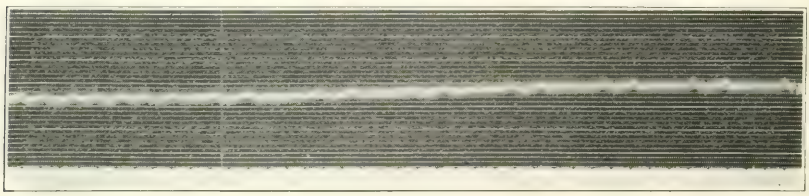


Fig. 1.—In this, as in the following figures, the time marker indicates fifths of a second, and an ordinate of 15 mm. is equal to 1 millivolt. Lead I. A marked slowing of the heart rate with no change in the height of P.

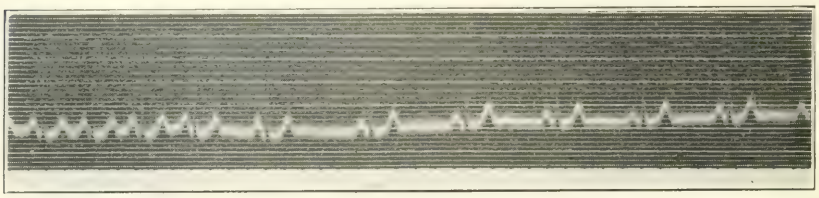


Fig. 2.—Lead II. The heart rate slows to less than one-half its usual rate. Coincident with this slowing, P becomes smaller and bifurcated.

feldt and Putzig[9] during expiratory slowing (Lead II). The last two observers found an increase in the height of the P complex in Lead I. Clinically, a decrease in the height of P has been noted during vagal pressure by Ritchie[10] (Lead II) and by von Hoesslin[11] (Leads I and

7. Einthoven, W.: Weiteres über das Electrokardiogramm, Arch. f. d. ges. Physiol., 1908, cxxii, 517.

8. Eyster, J. A. E., and Meek, W. J.: Cardiac Irregularities in Morphin Poisoning in the Dog, Heart, 1912-13, iv, 59.

9. Blumenfeldt, E., and Putzig, H.: Experimentelle electrokardiographischen Studien über die Wirkung der Respiration auf die Herztätigkeit," Arch. f. d. ges. Physiol., 1914, clv, 443.

10. Ritchie, W. T.: The Action of the Vagus on the Human Heart, Quart. Jour. Med., 1912-13, vi, 47.

11. von Hoesslin, H.: Beobachtungen über den Einflusz des Vagus auf das menschliche Herz, Deutsch. Arch. f. klin. Med., 1914, cxiii, 537.

II), and during forced respiration by Einthoven, Fahr and de Waart[12] (Leads I, II and III). In one of the cases reported by Einthoven, Fahr and de Waart, P became negative in Lead III and the same thing occurred in Lead I in one of von Hoesslin's and in one of Ritchie's cases.

A decrease in the height of P during vagus stimulation may be explained in the following ways:

Changes in the contractility of the auricles. Einthoven, Ritchie, and Eyster and Meek have regarded the flattening out of P as due to a diminution in the contractility of the auricles. Against this explanation in the present case is the fact that no change in the height of P occurred in the first lead although a well marked one occurred in the other leads taken immediately afterward. It is difficult to see, if the change in P is dependent on changes in auricular contractility, why it should be present in one lead and not in another.

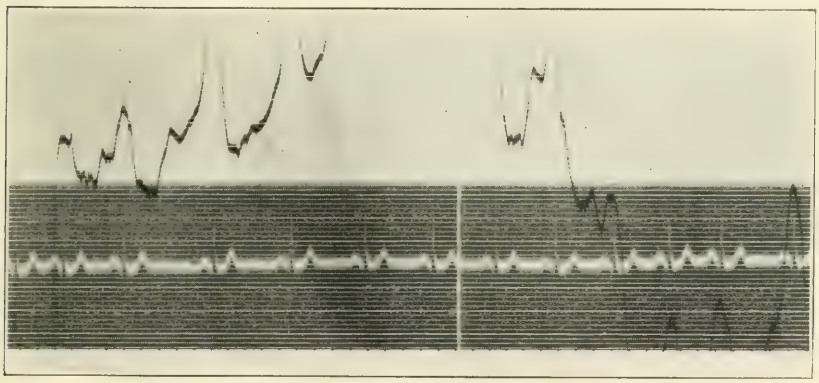


Fig. 3.—Lead III. Shortly after the beginning of expiration (upward movement of the venous pulse record) a marked slowing of the rate with flattening of P occurs.

Interference of two rhythms. Flattening out of P may be due to the simultaneous origin of two contraction waves in opposite portions of the auricle, so that their electrical effects partially or completely neutralize each other. Although this phenomenon occurred in the present case and will be described later, it is an improbable explanation where, as in the curves under discussion, the heart rate is constantly changing, for the rates of two centers would not be apt to change with equal speed. Furthermore, the deformed P complexes due to

12. Einthoven, W., Fahr, G., and de Waart, A.: Ueber die Richtung und die manifeste Grœsse der Potentialschwankungen im menschlichen Herzen und über den Einflusz der Herzlage auf die Form des Electrokardiogramms, Arch. f. d. ges. Physiol., 1913, cl, 275.

interference are followed by a shortened P-R interval and the P-R intervals of the complexes in question are normal.

Changes in the position of the heart. Einthoven, Fahr and de Waart have recently shown that although marked changes in the electrocardiogram may be produced by changes in the position of the heart, the changes in P which occur during forced respiration can not be due to this alone. It will be readily seen in Figure 4 that if a point in inspiration be compared with a point in expiration at which the distention of the lungs and consequently the position of the heart is the same, the heights of the P complexes differ widely. If the changes in P were due to changes in the position of the heart, the P complexes at these two points would be of equal height.

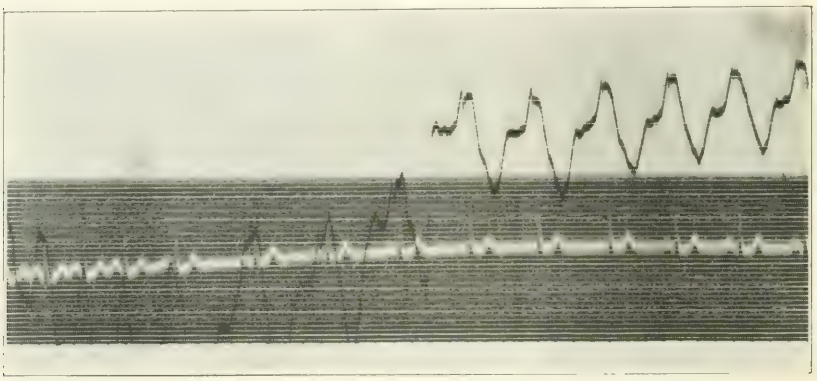


Fig. 4.—Lead III. The reduction in the size of P is so marked that it practically disappears.

Changes in the location of the pacemaker. Ritchie,[10] von Hoesslin,[11] and Einthoven, Fahr and de Waart[12] ascribed the inversion of P which they observed after vagal stimulation, to some change in the location of the pacemaker or to some change in the path of the contraction wave over the auricles. Lewis[13] has shown that a change in the shape of P indicates a change in the location of the pacemaker. He found for Lead II that if the impulse originates in the upper portion of the auricle, P is upright. When the impulse originates in the median portion of the auricle, P approaches the isoelectric state and when it originates in the lower portion of the auricle P is inverted. In the first lead the size and direction of P may not be markedly affected even when the impulse originates in the a-v node.[14] We

13. Lewis, T.: Galvanometric Curves Yielded by Cardiac Beats Generated in Various Areas of the Auricular Musculature. The Pacemaker of the Heart, Heart, 1910-11, ii, 23.

14. Hering, H.: Rhythmische Vorhofstachysystolie und Pulsus irregularis perpetuus, München. med. Wchnschr., 1914, lxi, 2057.

believe that in the present case the gradual flattening of the P complex in the second and third leads indicates that the pacemaker has moved downward. Since this occurred as the result of deep breathing, i.e., vagal stimulation, this explanation is also in accord with the experimental evidence that stimulation of the vagus displaces the pacemaker downward in the sinus node. The amount of change which P may undergo as a result of the migration of the pacemaker to the lower end of the sinus node has not yet been determined experimentally, so that it is impossible to tell whether this explanation would completely suffice in the present case where such marked changes in the size of P occurred.

RHYTHMIC AURICULOVENTRICULAR RHYTHM PRODUCED BY DEEP RESPIRATION

On January 25 it was noted that Patient 1 no longer showed a marked slowing of the pulse on deep respiration. Curves taken on this date disclosed an entirely different mechanism from that described above. Figure 5 is a typical example of these curves. Shortly after

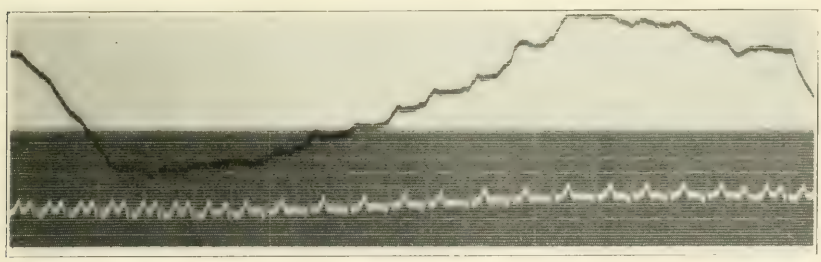


Fig. 5.—Lead II. Shortly after the beginning of expiration an atrioventricular rhythm appears. The P waves of this rhythm are inverted and the P-R intervals are shortened from about 0.15 seconds to about 0.09 second. The onset of the a-v rhythm is abrupt. The heart rate slows slightly at the beginning of this rhythm, but is faster than the heart rate during expiration in the previous figures. The inverted P complexes vary somewhat in form. The R complexes are unusually tall.

the beginning of expiration a slight slowing of the rate occurred. Coincident with this change in rate an abnormal rhythm appeared. This new rhythm was characterized by an inversion of P and marked shortening of the P-R interval. With the exception of a distinct increase in the height of R, the ventricular complexes were unchanged. These characteristics indicate that the site of origin of the new rhythm was in the upper part of the a-v node. The ectopic rhythm persisted for a short time and then the normal rhythm returned. This mechanism was repeated after each deep breath.

A very similar dislocation of the pacemaker could be produced in Case 2 also by forced respiration. In Figure 8, it will be seen that,

as in Figure 5, expiratory slowing is accompanied by the appearance of an abnormal rhythm. Here also the abnormal rhythm is characterized by inversion of P and shortening of the P-R interval indicating that the pacemaker was located in the a-v node. In this case the abnormal rhythm could also be produced by pressure on the right vagus in the neck. After the patient had been under observation for

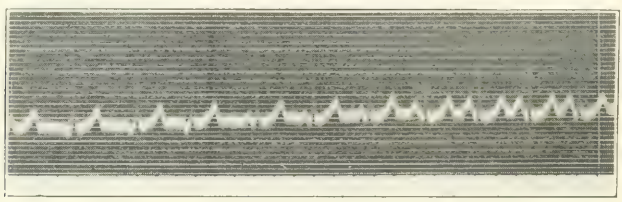


Fig. 6.—Lead II. Several transitional complexes occur at the end of an attack of a-v rhythm. The first two are diphasic, the third upright but small, and the fourth differs from the normal P complexes only in being more pointed. The P-R intervals of the first three transitional beats are shortened; that of the fourth is normal.

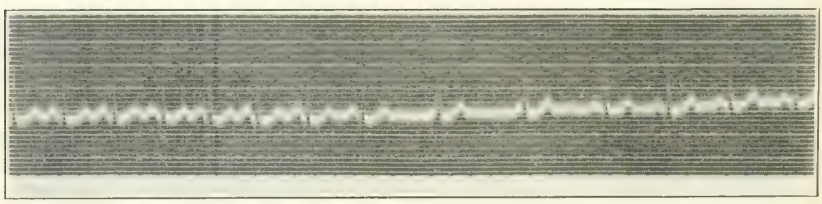


Fig. 7.—Lead III. The P and R complexes 9 and 10 are partially coincident. The P-R intervals are about 0.07 and 0.03 second, respectively. The P complexes are upright. The auricles responded to the sinus, the ventricles to the a-v node.

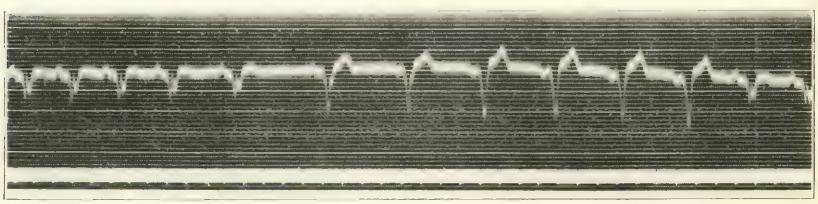


Fig. 8. Case 2. Lead III. Expiratory slowing is accompanied by the appearance of an abnormal rhythm. The first two Ps of this rhythm are diphasic, the rest are inverted. The P-R intervals are shortened from about 0.17 sec. to 0.1 sec. The ventricular complexes of the a-v rhythm are abnormal.

two or three days it began to appear spontaneously and at the end of a week was the rhythm usually present. During this period it could be abolished in favor of the normal rhythm by giving atropin subcutaneously in doses sufficient to paralize the vagi. In Case 2, the ventricular complexes during the abnormal rhythm, in contrast to

those of Case 1, are strikingly abnormal. As this modification of the ventricular complexes has no bearing on the subject treated in the present article, however, it will not be discussed here.

Transitions from a-v to normal rhythm and vice versa are of considerable interest. These are sometimes abrupt (Fig. 5). At other times they are more gradual, and between complexes of definite sinus origin and others of definite a-v origin two or more transitional complexes may occur (Figs. 6-8). The P deflections of these complexes are of two types, they are either upright and smaller than the P deflections of sinus origin or they are diphasic.

Two explanations may be offered for such transitional beats. They may be due to the interference of two contraction waves, the result of simultaneous impulse formation at the sinus and a-v nodes, or they may be due to a displacement of the pacemaker within the sinus node before it passes to the a-v node. The first explanation is the correct one according to Lewis[2] when the P-R interval is decidedly less than

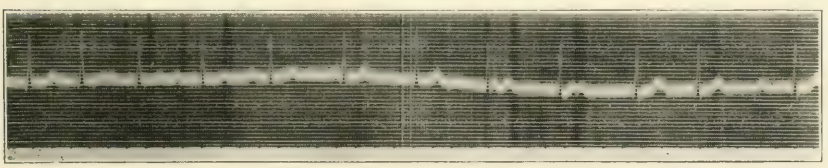


Fig. 9.—Case 3. Lead III. The P-R intervals of the earliest cycles are about 0.36 sec. From Cycle 5 to Cycle 10, inclusive, auricles and ventricles are contracting independently and the ventricles contract six times during this period while the auricles contract but five.

normal. This is true of the first three transitional beats of Figure 6. The last transitional beat of Figure 6 shows a slightly deformed P complex and a normal P-R interval so that it is possible that both these phenomena occur. Similar explanations may be offered for the fact that the inverted P complexes may vary considerably in size and form (Fig. 5). This may be due either to the interference of two rhythms or to changes in the location of the pacemaker within the a-v node.

RHYTHMIC ESCAPE OF THE VENTRICLES WITH ATRIO-VENTRICULAR DISSOCIATION PRODUCED BY DEEP BREATHING

The dislocation of the pacemaker which occurred in Case 3 during forced respiration was somewhat different from that observed in Cases 1 and 2. Figure 9 shows the effect of forced respiration on the heart mechanism in this case. The patient had a slight degree of heart block. The P-R interval was so long that P fell on T of the ventricular complex of the previous cycle when the heart was beating at its usual rate. At the beginning of expiratory slowing P

and T gradually separate (Fig. 9), but the P-R interval remains the same. After the fourth cycle of Figure 9, however, the P-R interval gradually becomes less and less until in the seventh cycle P falls on R. P then appears between R and T and finally falls on T in the tenth cycle. Thereafter the P-R interval is the same as at the beginning of the figure. Between Cycle 4 and Cycle 11, auricles and ventricles contracted independently. During the period of dissociation the ventricles were contracting more rapidly than the auricles and this difference in rate was so marked that the ventricles contracted six times while the auricles contracted only five. We are therefore dealing with the escape of a center located low down in the junctional tissues as the result of expiratory slowing of the sinus rhythm. The failure of the auricles to respond to the lower center was probably due to the heart block present. That short periods of dissociation may result when the idioventricular rhythm escapes as a result of sinus slowing even when there is no heart block is shown by Figure 7. This curve was obtained from Case 1. The P and R complexes of the first two cycles after slowing began are partially coincident. The P complexes are upright and so far as can be told are of the same form as those immediately following them. These characteristics indicate that the contractions which gave rise to these complexes were of sinus origin. The marked shortening of the P-R intervals makes it certain, however, that the ventricles responded to the junctional tissues. The dissociation of auricles and ventricles in this instance is due to the fact the stimulus from the center in the junctional tissues did not reach the auricles until they had become refractory.

In each of the cases described in this article, a-v rhythm could, at certain times, be produced at will by deep respiration. So far as we know, no similar cases have previously been reported. The production of a-v rhythm in man by deep breathing is analogous to the production of a-v rhythm in animals by stimulation of the vagus. In the clinical cases, however, in contrast to the experimental, the production of a-v rhythm is aided by a pathologic increase in the inherent rate of some center in the junctional tissues.

SUMMARY

Three cases are reported showing changes in the location of the pacemaker associated with deep respirations. These changes may be divided into three classes.

1. Migration of the pacemaker within the sinus node, or within its immediate neighborhood.

2. Migration of the pacemaker from the sinus node to the a-v node. The transitions from the normal rhythm to the a-v rhythm and

vice versa were sometimes abrupt, but at other times transitional beats occurred which were probably due to interference phenomena.

3. Escape of the idioventricular rhythm with complete atrioventricular dissociation, during which the ventricles contracted more rapidly than the auricles. This occurred both in a case in which there was a slight degree of heart block and in a case in which this complication was not present.

217 North Thayer Street.

THE SYMPTOMS OF URINOD POISONING*

FRANK A. HARTMAN, PH.D.
BOSTON

I. INTRODUCTION

The study of the toxic nature of urine has interested investigators for many years. The continued attraction which this subject affords is no doubt due to the part which urinary retention plays in the vital economy. When it occurs it may produce only headache and a general malaise, yet it cuts down the efficiency to a marked degree. In severer cases it may lead to grave symptoms and even death.

In attempting to account for this poisoning investigators have more often attributed it to a single urinary constituent. But a few have concluded that it was due to a number of substances.

In the study of a new substance[1] which is found in all urines, it was noticed that this substance, called *urinod,* produced headache and malaise. This led to a more careful investigation of its toxic nature.

From experiments conducted thus far, urinod seems to be one of the most toxic substances in normal urine. It is thought, therefore, that it may account for some of the symptoms of uremia. It must be understood, however, that this study does not attempt to explain uremia by the aid of urinod alone, but merely to show that it may be a contributing factor.

II. PREPARATION OF URINOD

Urine was treated with dilute sulphuric acid so as to make about 3 per cent. concentration of acid. The mixture was permitted to stand several days, becoming darkly colored and giving off disagreeable odors. At this stage the urine was distilled and the distillates were extracted with ether.[2] Acids, phenols and bases were removed from the ether extract by shaking successively with aqueous solutions of sodium carbonate, sodium hydroxid and hydrochloric acid. The ether solution, containing neutral substances, was concentrated and then subjected to steam distillation to remove urinod from the less volatile substances.

The steam distillation gave a low boiling thio-compound, urinod, a high boiling compound and sulphur in the distillate. This distillate was extracted with ether. The ether extract was washed with solutions of sodium carbonate, sodium hydroxid, and hydrochloric acid to remove possible traces of acids, phenols and bases. The ether solution was then dried with calcium chlorid and shaken in a separating funnel with metallic mercury to remove sulphur. The ether solution was concentrated and finally distilled in vacuo.

---

* Submitted for publication March 19, 1915.

* From the laboratory of chemistry of the University of Washington and the laboratory of physiology of the Harvard Medical School.

1. Dehn, W. M., and Hartman, Frank A.: Jour. Am. Chem. Soc., 1914, xxxvi, 2136.

2. For method of extraction see Dehn and Hartman, *loc. cit.,* Note 1, p. 2126.

The first distillate up to about 100 C. at 30 mm. always contained oil, with an allyl mustard oil or ethyl xanthate odor. The middle fraction, boiling at 108 C., with 29 mm. pressure, contained urinod, while the final fraction contained an oil boiling at 170 C. with 29 mm. pressure.

From the first seven hundred liters of distillate from 1,000 liters of urine 5 gm. of urinod were obtained. Of course some was lost during the handling, acidulation, distillation, ether extraction, concentration, treatment with acids and alkalies, distillation with steam, reextraction, drying and distillation in vacuo. It has been estimated that urinod occurs to the extent of one to two parts in 100,000 parts of urine.

III. PROPERTIES OF URINOD

The boiling point of urinod is 108 C. at 28 mm. It cannot be distilled at ordinary pressure without decomposition. Urinod does not solidify in an ordinary freezing mixture. It is a light yellow oil, slightly heavier than water. It is soluble in ordinary organic solvents, but is insoluble in water. It is very volatile with steam. It has a very penetrating, persistent, nauseating odor of urine. It is very toxic, as will be shown in the experiments to follow.

Urinod darkens rapidly in direct sunlight, reduces solutions of potassium permanganate and ammoniacal silver nitrate in the cold, reacts with Millon's reagent, but not with Fehling's reagent. With fixed alkalies the odor of urinod is changed to a terpene-like odor.

IV. COMPOSITION[3] AND STRUCTURE OF URINOD

The empirical formula of urinod has been established as C_6H_8O.

Urinod is insoluble in hot dilute solutions of hydrochloric and sulphuric acids; therefore it cannot contain a basic (or alcohol-oxygen or ether-oxygen) group.

Urinod is optically inactive, hence, if not a racemic mixture, it cannot contain an asymmetrical carbon atom.

Urinod reacts with bromin contained in carbon tetrabromid, giving a strong evolution of hydrogen bromid and a solid bromo-derivative; hence it is a *cyclic* compound. Additional evidence of its cyclic nature is obtained from the formation of a dinitro-derivative (golden needles melting at 78 C.) by treatment with cold, dilute nitric acid.

Urinod reacts with semicarbazide, forming a compound melting at 254 C. Therefore it is either a *ketone* or an *aldehyde*.

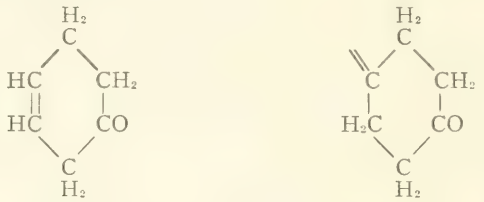
Urinod reacts with hydroxilamin phenylhydrazin and hydrogen sulphid, forming derivatives not possessing the characteristic odor of urinod. These compounds indicate the presence of a *carbonyl* group in urinod.

The oxidation of urinod by ammoniacal silver nitrate, also by aqueous solutions of potassium permanganate, indicates close relationship with hydrobenzene derivatives.

3. Dehn and Hartman, *loc. cit.*, Note 1, p. 2138.

No known compounds having the same empirical formula as urinod possess the same properties.

From the foregoing considerations and other considerations, discussed in a paper[4] previously published, two possible structural formulas have been obtained.



That urinod may possess the second formula gathers support from consideration of its characteristic toxic properties and its odor—the isocyanids and other bivalent[5] carbon compounds being both toxic and malodorous.

V. URINOD OCCURS IN THE CONJUGATED FORM

That urinod does not occur free in fresh, normal urine is concluded from consideration of the following evidence:

1. The odor of urinod is absent from fresh urine.
2. Hydrolysis by fermentation or chemicals develops composite odors containing the odor of urinod.
3. Urinod is very toxic; therefore for physiological reasons it must be rendered harmless by conjugation.
4. Although urinod is easily volatile with steam, each fraction of the distillate from urine and even the heated residue gives off the odor of urinod, hence not only is its formation by hydrolysis difficult, but, *a priori,* it must exist in the conjugated form.
5. Additional support is given to this conclusion from a consideration of the many other conjugated substances in the urine.

VI. METHODS OF ADMINISTERING URINOD

At the beginning of the experimental work with urinod, a method of injection was sought. But for the reason that solvents for urinod are more or less harmful to the body tissues, this method was thought to be rather unsatisfactory.

In a few cases the pure oil was injected intramuscularly, intravenously or subcutaneously. The use of a syringe was found to be too wasteful because of the oil adhering to it; therefore in the cases

4. For more detailed discussion see Dehn and Hartman, *loc. cit.,* p. 2141.

5. Nef: Ann. d. Chem. (Liebig's), 1894, cclxxx, 303; 1895, cclxxxvii, 274; 1897, ccxcviii, 202. Wade: Jour. Chem. Soc., 1902, lxxxi, 1596. Lawrie: Am. Chem. Jour., 1906, xxxvi, 487.

of intramuscular and subcutaneous administration the urinod was dropped from an ampoule into an incision in the muscle or skin.

The method used most often in animal experimentation was to drop the urinod into the pharynx. The capillary tube of a weighed ampoule of urinod was broken and the oil forced out by heat. The weight of the ampoule containing the remainder of the urinod together with its broken capillary, subtracted from the original weight, gave the amount of oil used.

In the two experiments on man in which urinod was taken intentionally, an open vial containing a drop of urinod was used. One nostril was held shut, the other was placed in contact with the opening of the vial, so that enough space was left for the inhalation of outside air. In this manner urinod vapor was mingled with the inspired air.

Because of the difficulty in obtaining urinod in any considerable quantity, the use of small animals was necessary in a majority of cases. In the following experiments something less than a total of 300 milligrams was used.

VII. EXPERIMENTAL RESULTS

1. The frog.—Urinod was quite toxic for frogs, one five-hundredth to one eight-hundredth of the body weight being sufficient to produce death in from twenty-five minutes to a few days, depending on the place of injection. Injections in the muscles of the hind leg were least effective, while doses by the mouth were quickest in their action. Death was due to the stopping of respiration.

A number of symptoms developed in the cases in which death did not take place for several hours. Twitchings and dyspnea were followed by convulsions and a high degree of irritability. In the last stages the animal became limp and would not respond to stimulation.

It is an important fact to be borne in mind that the heart was unaffected by urinod. The heart finally ceased beating, due to asphyxia. The reason that it continued so long after the lungs were inactive was no doubt due to respiration through the skin.

Tree frogs were used in the following experiments because of their small size.

A. A frog was given urinod by the mouth in the proportion of 1 part urinod to 460 parts body-weight. The frog jumped about a few times and then remained quiet. After 2.5 minutes the heart beat 100 times per minute; there were two or three jerky inspirations and then a long pause. After five minutes the animal was so weak that he could not sit up; breathing had practically stopped. After seven minutes he would kick very weakly when disturbed; he jerked and twitched much of the time. After ten minutes convulsions occurred. After thirteen minutes the heart had been reduced to 44 beats per minute; at this time tremors passed over the body. The heart continued beating for twelve minutes longer.

In this animal a condition of non-irritability had developed after ten minutes while death occurred in twenty-five minutes. The intermediate symptoms, such as twitchings and convulsions, were scarce and short-lived. Evidently more than the lethal dose had been given.

An examination of the esophagus and stomach showed that a large proportion of the urinod had not been absorbed. This is not surprising considering the insolubility of urinod in water.

B. Another frog was given urinod in the proportion of 1:800 body weight, by dropping the oil into an incision in the abdominal wall. The frog was very frantic at first, jumping about and in various other ways showing his discomfort. After six minutes breathing had become slow and jerky. After sixteen minutes convulsions occurred, preceded by jerking and twitching; his legs were sprawled out and he was so weak that, although responding to very slight stimulation he could barely jerk. After thirty-six minutes the heart beat 72 times per minute; there was congestion of the blood vessels on the abdomen. After fifty minutes there was a quiet period interrupted by occasional jerks and twitches. After 1.75 hours, the heart beats were only reduced to 70 times per minute; the frog was no longer nervous, and breathing had become more spasmodic than ever. After three hours the heart beats were 56 per minute and twitching of individual muscles continued. After 4.5 hours the frog stopped breathing. At 5.5 hours after the administration of urinod the heart stopped beating.

This experiment demonstrates that although urinod is practically insoluble in water it is absorbed in sufficient quantity to affect the respiratory center. Very little, apparently, is necessary to cause dyspnea, as in six minutes this occurred, while it took four and one-half hours to carry enough of the poison to this center to stop its action completely.

As in the preceding case, very little of the urinod given was carried away.

The decrease in the rate of the heart beat is always in proportion to the decrease in the respiration and lags behind, as these experiments and those that follow demonstrate. Moreover, the fact that urinod has no direct action on the frog's heart was shown in another way: Excised frog-hearts were immersed in Ringer's solution, in which a drop of urinod had been placed. The rate of beat was then compared with other hearts in the same conditions except that urinod was absent. With ten trials no effect could be noticed even when urinod was brought in contact with the heart in the form of a large drop.

C. In a third experiment[6] urinod in the proportion of 1:2,500 body-weight had been injected into the muscles of the thigh of a frog. He immediately showed his irritation by jumping about. (These first effects were undoubtedly due partly to the mechanical irritation in making the incision.) After two minutes he jumped about very frantically, but his efforts were very weak. After three minutes he became very quiet; the injected leg had become paralyzed. After thirteen minutes he frequently gaped his mouth. After eighteen minutes the heart beat 59 times per minute and the muscles occasionally twitched. After twenty-one minutes there were convulsions. After twenty-three minutes his breathing had become irregular and he had become very frantic. This was followed seven minutes later by a state of nonirritability. After thirty-two minutes breathing had all but ceased, muscular twitchings and heart beats being the only indications of life for a time. After thirty-five minutes there were six respirations in one minute, first a single respiration, then a pause of forty seconds, followed by 5 respirations in as many seconds; another pause then a few more respirations followed. The injected leg was by this time congested with blood. Judging by previous experiments, it was thought that these symptoms were ushering in the end. After seventy minutes, however,

6. It is worth while to call attention to the similarity of urinod poisoning to carvone poisoning, as carvone can be easily obtained, while urinod is obtained only after considerable time and labor. The chemical structures and physical properties of the two substances are somewhat similar. Carvone is an unsaturated cyclic ketone with a penetrating odor. It is insoluble in water. Although not so toxic as urinod, carvone produces death in the same manner by injury to the respiratory center. The heart stops beating only after asphyxiation. The preliminary symptoms are twitching and dyspnea, while in grave cases convulsions and conditions of non-irritability follow. These effects were studied in the frog, the lizard and the mouse.

the frog began to recover. After two hours he had apparently recovered. In three days the injected leg was red and swollen to the tip of the toes; the frog had again become quite sick. He died during the third day.

In this experiment it was not positively known whether death was due primarily to urinod or to secondary causes. The last symptoms preceding death were a gradually increasing weakness, with an increasing condition of non-irritability.

2. The lizard.—The animals used in these experiments did not belong to a very active species (*Gerrhonotus multicarinatus*).

The symptoms resulting from urinod poisoning resembled those present in the frog, except that there was considerable jerking of the body from side to side. Death was due to a checking of respiration. Urinod was administered orally in all cases.

A. A lizard was given urinod in the proportion of 1:14,000 body-weight. After two minutes the lizard began to jerk his body and twitch his eyelids. Gradually the spasms increased in vigor and breathing became more difficult. In the course of an hour the jerks became weaker. After 1.5 hours the animal lay still except with an occasional twitching. He would not move when stimulated. During the course of three hours, however, this state of nonirritability had gradually passed away, the animal showing complete recovery.

This experiment demonstrates that in urinod poisoning the symptoms may become very severe, even so far as prolonged paralysis; yet they may be followed by complete recovery. As soon as absorption of the poison commences, detoxification probably begins. If urinod does not occur in sufficient quantity, enough of that being absorbed can be rendered harmless so that a fatal poisoning does not result.

B. Urinod was given to another lizard in the proportion of 1:8,200 body-weight. In a little more than two minutes he began to twitch. The twitches gradually grew into jerks. After four minutes he crawled about slowly, jerking continually. After seven minutes he was so frantic that he darted first one way and then the other. After twenty minutes the ability to crawl had been lost, although there was continued jerking from side to side and twitching of the individual muscles. When placed on his back the lizard could not turn over. In the course of three hours he began to regain the use of his limbs. After four hours he had gained enough to right himself when placed on his back. However, he still continued to jerk. After nine hours he had recovered.

This experiment corroborates the results of the preceding one.

C. In a third experiment the dose of urinod was increased in order to study the symptoms leading up to death. One part urinod to 1,800 parts body-weight, was given. After the poison began to affect him the symptoms became very severe in a short time.

At six minutes he had begun to twitch, while in ten minutes the eyes were half shut and he was unable to crawl. He gradually grew weaker until at one hour from the time of administering urinod, the feet were sprawled out with the soles up. After 1.25 hours, the state of nonirritability was complete, twitching being the only sign of life. Death occurred 1.5 hours after urinod was given.

D. In the fourth case the dose of urinod was slightly increased beyond that used in the previous experiment.

Twitching of the eyelids followed a few seconds after administration. Spasms and paroxysmal breathing soon ensued. After ten minutes the limbs were so weak that they could not support the body. At the end of fifteen minutes an oily excretion was defecated.[7] This contained a large amount of urinod. By this time the eyes of the lizard were kept half closed and when picked up the

7. A lizard which was given a lethal dose of the oil carvone excreted a great deal of it six minutes afterward by the intestinal path.

animal was limp. In thirty-five minutes movement was reduced to occasional twitchings. Death occurred one hour after administering urinod.

Autopsies of animals killed by urinod have always shown a large amount of the poisonous oil in the stomach and intestines. Apparently there was always an attempt to remove the urinod by the intestinal path. In this experiment a considerable amount of the oil administered was removed from the body in this manner, but enough remained to kill. That death does not occur very rapidly, as with many soluble poisons, is undoubtedly because urinod is absorbed so slowly.

3. The mouse.—It was desirable to study the severer symptoms of urinod poisoning in mammals. Therefore, on account of the small amount of the poison required to produce these results, the mouse was used.

A mouse weighing 10.9 gm. was given urinod at intervals of a few hours to a few days, a drop being placed on the tongue.

First dose: 14.2 mg. (Ratio 1:767 body-weight.) The mouse immediately rubbed his snout on the floor of his cage, causing the oil to run out of his mouth. After two minutes he opened and closed his eyes, and then kept them half shut. After three minutes he jumped and twitched. After six minutes slow and difficult breathing developed. After ten minutes he ran about frantically. After twelve minutes he lay as though in a stupor. After fifteen minutes his eyes were open again. After seventeen minutes he ran about chewing things but not eating. In one hour his symptoms became milder. After two hours he twitched his ears continually. After three hours his body still twitched and he did not move even when stimulated in a manner which had previously provoked a response. After thirteen hours he trembled a great deal; his appearance was depressed.

He twitched for twenty-four hours with eyes half shut and breathing labored; he also seemed to be chilly. A few hours after the administration of urinod the fur became so moist from perspiration that the hair was matted as though it had been immersed in water.

Second dose: Six days after the first dose, the same mouse was given 8.2 mg. of urinod (1:1,329 body-weight). The symptoms, as described above, with little variation, again appeared.

Third dose: Eight minutes after the second dose, 10.3 mg. of urinod was given. In four minutes the mouse's eyes were completely shut; he twitched spasmodically. After six minutes he blinked his eyes. After eight minutes he ran frantically back and forth, occasionally rubbing his snout on the cage; his breathing was slow and irregular. After twenty-two minutes he was breathing rapidly again but irregularly; he also jerked his body. At thirty-two minutes there was a slight spasm. At thirty-seven minutes there was another slight spasm. After forty-two minutes he was frantic and possessed a high degree of irritability. After forty-seven minutes he was so irritable that he jumped at the slightest disturbance. At sixty minutes he had convulsions; then intervals of frantic running about with periods of quiet between them continued for several minutes. After nine hours he still trembled.

Fourth dose: Seven days after the first dose, 3 mg. of urinod was given. The mouse became nervous and twitched. This was followed by a period of quiet.

Fifth dose: Thirteen days after the first dose a drop of urinod was given at intervals during the course of thirty minutes until 60 mg. had been taken. (Ratio 1:182). In twenty minutes after the first drop was given the breathing was slow and difficult; the eyes were half shut and the mouse paid no attention to noises. After twenty-seven minutes the eyes were closed and the breathing was not only slow, but very irregular; first, there was a labored respiration, then two or three respirations followed rapidly. At twenty-eight minutes his head sank to the floor and his body jerked. After thirty-seven minutes he passed

into a state of nonirritability, breathing in paroxysms. At forty minutes he had a convulsion. At forty-five minutes convulsions again occurred. After one hour the breathing became more regular. After 3.5 hours the breathing was almost normal, but the body trembled. At 17.75 hours he was continually blinking his eyes, first one and then the other; there was also twitching of the ears. After nineteen hours he occasionally jerked. He kept up the blinking of his eyes until the end of the fourth day. In this movement he always lacked coordination as the eyes were blinking alternately instead of in unison. During this period he kept his eyes partly closed when not in the act of blinking. On the fifth day he had about regained his normal condition.

The symptoms developed in the mouse were muscular twitchings, heightened irritability, apparent drowsiness, labored paroxysmal breathing and dyspnea.

The same proportion of urinod was not as effective in the mouse as in the lizard for it was impossible to place the oil far enough into the throat to prevent removal to the floor of the cage.

4. Man.—Although the graver symptoms of urinod poisoning cannot be determined experimentally in man, he serves as an excellent subject for the study of many minor symptoms.

It was while working with urinod in the laboratory that its toxic nature was discovered through the effect produced on breathing its vapors. This was noticed whether working with the pure substance or with the complex distillate from acid-treated urine.

One of the first effects noticed was the intense nausea produced when working over small amounts of urinod. This was almost invariably followed by headache and mental depression. The headache was often occipital in its location.

During periods when I was handling urinod every day for a number of days together, I noticed loss of appetite and a heaviness of the stomach after eating, also constant weariness and frequent periods of drowsiness. Sometimes there was considerable irritability of temper. Occasionally there were marked restlessness and insomnia, although these were usually followed by heavy sleep.

The breathing of urinod from a small vial for ten minutes caused a peculiar sensation in the head, followed by a numbness in the occipital region. It became impossible to concentrate the attention for a time. Headache and depression soon developed. These symptoms, together with irritability of temper lasted for about five days.

At various times when under the influence of urinod I noticed an unusual chilliness. During one period, about three days in length, after I had handled more urinod than usual, I felt a great desire to micturate several times at night although the bladder might be practically empty. The urine gave a burning sensation in its passage. Moreover during this period some itching of the skin occurred. It may be mentioned that after long exposure to urinod, fatigue developed very early upon exertion.

In distilling urinod on one occasion, the stopper to the flask being accidentally removed, I inhaled a considerable quantity of the vapor. In a few seconds I realized a very peculiar, indescribable sensation in both the lumbar and occipital regions, the latter predominating. Shortly after it took effect I felt extremely restless and irritable. This feeling gradually passed off into headache and depression which lasted for a few days.

It was interesting to note the effect of urinod on other individuals. Those who remained for a few minutes in the room where urinod was being used, complained of headache and depression.

One individual inhaled urinod from a vial for five minutes. An extremely nauseating sensation was experienced while breathing the urinod and for sometime later. Headache began in a few minutes, with a tendency to sleep. The next day this person was extremely irritable. The effect passed away in about three days.

If one considers the minute trace of urinod which would be taken into the system by inhalation, one can realize how toxic the substance must be to produce the results described.[8]

An important characteristic of this toxic effect is the slowness with which it disappears.

The symptoms commonly shown by urinod poisoning in man are, nausea, headache (more pronounced in the occipital region), mental depression, loss of appetite, heaviness of stomach after meals, constant weariness and drowsiness. The symptoms occasionally developed are, irritability of temper, marked restlessness, insomnia, inability to concentrate the attention, chilliness and desire for frequent micturition.

VIII. THE LETHAL DOSE OF URINOD

The quantity[9] of urinod necessary to produce the graver symptoms, or even death, is very small, judging from the experiments on animals.

The amount given them was small, but the portion actually absorbed or carried away by the circulation was very minute, as shown by the quantity remaining unabsorbed. In postmortem examination, urinod given by the mouth is largely recovered in the stomach and intestines, while that injected intramuscularly or subcutaneously is found little diminished in that region.

From these facts it is very certain that amounts considerably smaller than 1 part to 800 parts body-weight (frog) would produce death if it were all absorbed in the circulation. Moreover, if other substances that are normally eliminated by the kidneys, accumulated in the blood, the resistance of the tissues would probably be diminished.[10] It is also quite possible that there is an increase in urinod under certain conditions.

---

8. Later experiments have shown that when all of the inhaled air is made to pass over urinod in the following apparatus only 2.8 mg. are volatilized in thirty minutes. The following arrangement was used: A liter bottle was supplied with dry air from a tower of $CaCl_2$ and soda lime. A short wide-mouthed vial, containing a weighed quantity of urinod, was supported so that the urinod was about 0.5 cm. below the opening of the exit tube of the bottle. This exit tube was connected with a piece of rubber tubing. The rubber tubing fits closely in one nostril, the other being held shut during inspiration. At expiration the rubber tube was pinched to prevent any back flow of air. In this way the amount of urinod used could be accurately determined.

9. It has been found recently that by dissolving urinod in olive oil it is more toxic when injected subcutaneously. When given thus, one part of urinod to 3,000 parts body-weight is sufficient to kill mice; even then only about one-half to one-third of the oil is absorbed.

10. Cawadias, Alexandre (Compt. rend. Soc. de biol., 1910, lxix, 153) working with blood serum of uremic patients adopted the hypothesis that a physicochemical modification of the colloids in the serum made an increase in the toxicity of substances accumulating in uremia — the accumulation of potassium salts, ammonium carbonate and urea played a part in this modification of the colloids.

IX. INDICATIONS OF URINOD RETENTION IN THE BODY

Because of the effect which it might have, on account of its toxic nature, it is of interest to note the indications of urinod-retention in the body. Urinod gives the characteristic odor to urine; therefore its presence is recognizable from its odor. Also, urinod is a normal constituent of all urines. If at any time urine does not have an odor, from the above considerations it would be concluded that urinod had been retained in the body. The odor of urine is said to be absent in some cases of uremia,[11] therefore, in those cases urinod is probably retained in the body.

Positive evidence of urinod retention is obtained from the fact that the skin and breath of uremic patients are often described as having a urinous odor.[12] This observation and the following would indicate that not only was urinod retained but that it was present in the free condition, as it is the unconjugated substance which gives the urinous odor.

Christison's[13] observation is very important in its bearing on this point. He found the odor of urine in the heart blood of a man who had died of uremia. Two ounces of blood were removed with great care from the heart so that contamination was impossible. This blood was shaken with alcohol and filtered. Following this it was carefully evaporated to dryness on a vapor bath. The residue was then treated with HNO_3, giving the odor that is obtained when urine is treated with HNO_3.

X. A COMPARISON OF URINOD SYMPTOMS WITH UREMIC SYMPTOMS

The symptoms of urinod poisoning, as shown in the foregoing experiments, are headache, nausea, loss of appetite, heaviness of the stomach after meals, twitching, irritability of temper, mental dulness, physical weariness, drowsiness, dyspnea, convulsions and a condition of nonirritability. Occasionally there are other symptoms such as restlessness, insomnia, itching of the skin, frequent micturition, chilliness and fatigue after slight exertion.

Tyson[14] says that a frequent desire to micturate is an early symptom of uremia. In regard to the symptom of chilliness, that was described by Saundby.[15] Fatigue after slight exertion was observed in a case of uremia by Foster.[16] All of the other symptoms of urinod

11. de Beauvais, M.: Compt. rend. Acad. d. sc., 1850, xlvii, 641.
12. Willson: Jour. Am. Med. Assn., 1905, xlv, 23; Porter, W. H.: Renal Diseases, 1887, p. 84; Christison, R.: Granular Degeneration of the Kidneys, 1839, p. 202.
13. Christison, R.: Granular Degeneration of the Kidneys, 1839, p. 169.
14. Tyson, James: Bright's Disease and Diabetes, 1881, p. 103.
15. Saundby, Robert: Lectures on Renal and Urinary Diseases, 1896, p. 158.
16. Foster, N. B.: The Archives Int. Med., 1913, xii, 455.

poisoning are often present in uremia[17] with one exception, and that is the paralysis of the center which coordinates the movement of the eyelids (observed in one instance in the mouse). So far I have not found a description of this symptom in uremia.

An important organ which is affected in uremia is the heart. Urinod has no effect on this so far as experiments up to this time have shown. The changes in blood pressure which sometimes are present in uremia apparently bear no relation to the retention of urinod, as it has been impossible to obtain any effect on blood pressure by the use of urinod.

Apparently urinod plays no part, or at most very little, in the production of dropsy, as only in one instance was there any appearance of edema.

Therefore it is concluded from the above considerations that the symptoms of urinod poisoning resemble very much the nervous symptoms of uremia. It is possible, then, that urinod is the cause of some of the nervous symptoms present in uremia. On the other hand, there are other substances in the urine which produce symptoms such as twitching, convulsions, and a condition of nonirritability and coma. For this reason it is probable that these substances contribute to this effect on the nervous system. A varying amount of these substances which influence the nervous system might account for the variations in uremic symptoms. Obermayer and Popper[18] thought that the changing clinical picture in uremia might be due to the changing power of elimination in the kidney for different substances.

SUMMARY

1. Urinod[19] is prepared from the distillates of acid-treated urines. It is a neutral malodorous oil boiling at 108 C. with 28 mm. pressure. It is a cyclic ketone with the empirical formula C_6H_8O.

2. The symptoms produced by urinod ordinarily are: nausea, headache, loss of appetite, heaviness of the stomach after meals, twitching, irritability of temper, mental dulness, physical weariness, drowsiness, dyspnea, convulsions and a state of nonirritability.

3. Urinod appears to be one of the most toxic substances in urine

4. Cases are cited in which there have been indications of urinod retention in the body.

5. The symptoms of urinod-poisoning resemble the nervous symptoms of uremia. Urinod retention, therefore, might partly account for these nervous symptoms.

17. Fürbringer, Paul: Diseases of the Kidneys, Trans. by W. H. Gilbert, 1895, i, 38; also Garrod, A. E.: Osler's Modern Medicine, 1909, vi, 91.

18. See Friedrich and Hugo: Ztschr. f. klin. Med., Berl., 1911, lxxii, 332.

19. Further studies are being made on urinod.

A STUDY OF THE SEVERAL FACTORS OF ACID EXCRETION IN NEPHRITIS *

WALTER W. PALMER, M.D. †
BOSTON
AND
LAWRENCE J. HENDERSON, M.D.
CAMBRIDGE, MASS.

Recently acidosis in nephritis has attracted much attention among clinical investigators and many articles dealing with the subject have appeared. Considerable evidence has accumulated which leaves little doubt that in certain of the more severe types of chronic renal disease there occur mild grades of acidosis. The facts revealed in various researches which confirm the frequently observed clinical picture of acidosis in nephritis include lowered alveolar carbon dioxid tension, the decreased affinity of hemoglobin for oxygen, reduced alkalinity of the blood, increased intensity of urinary acidity (hydrogen ion concentration) and the retention of alkali by the body in cases in which the kidney is capable of the rapid elimination of an excess of alkali.[1]

The purpose of the present communication is to present certain data on the acid excretion in nephritis which we have collected over a period of three years during our investigation of the process of acid excretion in health and disease.[2]

---

* Submitted for publication April 8, 1915.

* From the Wards and Chemical Laboratory of the Massachusetts General Hospital.

† Henry P. Walcott Fellow, Harvard Medical School, 1912 and 1913. House Physician, Massachusetts General Hospital, since 1913.

1. Palmer, Walter W., and Henderson, Lawrence J.: Clinical Studies on the Acid Base Equilibrium and the Nature of Acidosis, The Archives Int. Med., 1913, xii, 153; Sellards, Andrew W.: The Essential Features of Acidosis and Their Occurrence in Chronic Renal Disease, Bull. Johns Hopkins Hosp., 1914, xxv, 141, for a review of the literature on this subject.

2. Henderson, Lawrence J., and Palmer, Walter W.: The Intensity of Urinary Acidity in Normal and Pathological Conditions, Jour. Biol. Chem., 1913, xiii, 393; The Extremes of Variation of the Concentration of Ionized Hydrogen in Human Urine, Jour. Biol. Chem., 1913, xiv, 81; Newburgh, L. H., Palmer, Walter W., and Henderson, Lawrence J.: A Study of Hydrogen Ion Concentration of the Urine in Heart Disease, The Archives Int. Med., 1913, xii, 146; Palmer, Walter W., and Henderson, Lawrence J.: Clinical Studies on Acid Base Equilibrium and the Nature of Acidosis, The Archives Int. Med., 1913, xii, 153; Henderson, Lawrence J., and Palmer, Walter W.: The Several Factors of Acid Excretion, Jour. Biol. Chem., 1914, xvii, 305; Henderson, Lawrence J., Palmer, Walter W., and Newburgh, L. H.: The Swelling of Colloids and Hydrogen Ion Concentration, Jour. Pharm. and Exper. Therap., 1914, v, 449.

I. MATERIAL AND TECHNIC

Our observations include the study of fifty-eight cases of varying grades of nephritis for a total of 377 days. As in our studies on normal individuals, we have determined for the twenty-four hour amount of urine the hydrogen ion concentration, acid excretion and ammonia excretion in all cases.

In eleven of the subjects, however, the urinary nitrogen and noncoagulable nitrogen in the blood are added to the observations. Certain other facts of clinical interest, such as phenolsulphonephthalein excretion, blood pressure, specific gravity, albumin and the microscopical examination of the urinary sediment, are also included among the data. We have made no attempt to carry out elaborate studies in metabolism, although recognizing the value of such a procedure, but have, anticipating criticism, studied the effect of diet on the acid factors of the urine in two normal individuals, using the same articles of food which all the cases we report received while under observation. The study of the effect of diet in the two normal subjects is divided into three periods. In the first period a low protein diet was obtained by the use of soluble arrowroot starch as a principle article of food, while in the second period we employed essentially the hospital "nephritic diet." The high nitrogen in the third period was secured by the ingestion of large quantities of milk and eggs.

The methods of study are the same as those described in our earlier papers. Urinary nitrogen and noncoagulable nitrogen in the blood were determined according to the methods of Folin.[3] All observations were made in duplicate. It may not be out of place here to call attention to certain precautions necessary in the study of acid excretion. First, it is highly desirable, in fact very necessary, to have a special nurse to control the diet and insure proper collection of the urine. An approximately standard diet should be used in all cases, in order to permit reliable comparisons. For a diet varying markedly from the one used in our work (Table 1) normal controls should be secured. Blatherwick[4] and others have shown that by proper selection of diet in reference to the acid or basic nature of its ash, the acid factors of the urine may be markedly influenced.

Such extremes in diet practically amount to the feeding of acid or alkali, experiments with which we have already reported.

It is absolutely essential that the urine be collected in clean receptacles and well preserved. We have found chloroform with 5 per cent.

3. Folin and Farmer: Jour. Biol. Chem., 1912, xi, 493; Folin and Denis: Jour. Biol. Chem., 1912, xi, 527.

4. Blatherwick, N. R.: The Specific Rôle of Foods in Relation to the Composition of the Urine, The Archives Int. Med., 1914, xiv, 409.

thymol, 10 c.c. to a five-pint bottle, to be quite satisfactory. During the collection of the twenty-four hour amount, the bottle should be kept well stoppered in a cool place, preferably in an ice chest, or if this be not practical, the bottle must be vigorously shaken after the

TABLE 1.—Normal Subjects, Varying Amounts of Protein

Volume, c.c.	Hydrogen Ion Concentration	Acid N/10 c.c.	Ammonia, N/10 c.c.	Acid + Ammonia	Acid ÷ Ammonia	Ammonia N, Gm.	Total Nitrogen, Gm.	Ammonia Nitrogen Ratio	Diet
W. W. P.									
1,720	5.9	232	395	627	0.59	0.55	10.3	5.3	Soluble starch, cream, sugar, fruit, potato, butter, lettuce.
2,085	6.2	196	373	569	0.53	0.52	8.9	5.9	
1,405	6.0	258	302	560	0.85	0.42	6.6	6.4	
1,300	5.7	276	320	596	0.86	0.45	6.2	7.3	
2,349	5.7	392	560	952	0.70	0.76	16.0	4.8	Fruit, cereals, bread and butter, cream, sugar, eggs, potato, macaroni, milk, cheese.
1,687	5.5	451	475	926	0.95	0.67	14.0	4.8	
935	5.4	475	503	978	0.95	0.70	10.8	6.5	
1,005	5.5	345	434	779	0.80	0.61	10.6	5.7	
2,105	6.0	370	595	965	0.62	0.84	20.4	4.1	Same as in Period 2 except marked increase in eggs.
2,197	5.7	495	654	1,149	0.76	0.92	22.0	4.2	
2,088	5.3	720	785	1,505	0.92	1.10	24.9	4.4	
1,652	5.3	760	842	1,602	0.90	1.18	23.2	5.1	
Average acid-ammonia ratio is 0.785.									
J. H. M.									
1,310	5.4	257	320	577	0.80	0.45	8.7	5.2	Same as in Subject W. W. P.
2,447	6.1	191	275	466	0.70	0.39	7.0	5.6	
1,545	6.0	181	236	417	0.77	0.33	5.4	6.1	
1,460	6.3	139	245	384	0.57	0.34	4.5	7.5	
2,043	5.5	300	370	670	0.81	0.52	12.0	4.3	Same as in Subject W. W. P.
945	5.7	316	332	648	0.95	0.47	9.1	5.2	
993	5.7	330	336	666	0.98	0.47	9.8	4.8	
1,322	5.6	303	360	663	0.84	0.51	9.4	5.4	
1,600	6.2	298	440	738	0.68	0.62	13.9	4.5	Same as in Subject W. W. P.
2,138	5.9	368	457	825	0.81	0.68	17.2	4.0	
1,925	5.4	595	660	1,255	0.90	0.93	20.5	4.5	
1,462	5.3	535	716	1,251	0.75	1.00	17.2	5.8	
Average acid-ammonia ratio is 0.797.									

addition of each single specimen. The acid factors should be determined soon after the completion of the twenty-four hour amount. Especial care should be taken to exclude alkalies or acids from all drugs used, not only during the observations but for some time before starting the experiment.

Subjects with infections of the genito-urinary tract are wholly unsuitable because of the activity of bacteria in changing rapidly and markedly the relations between acid and ammonia, as well as the hydrogen ion concentration of the urine. Female patients are less satisfactory than males because of the frequency of cystitis and the difficulty in securing uncontaminated specimens. However, with the divided bed pan devised by Dr. Denis of the Massachusetts General Hospital chemical laboratory, satisfactory collections may be made.

II. CONTROLS

The experiments on the two normal subjects, W. W. P. and J. H. M., were so planned as not only to include the usual variation in diet which might occur in the pathological cases, but also the unusual extremes of high and low protein intake, while using in the two cases, as far as possible, the same relative amounts of the different articles of food. The results of these experiments may serve as a standard for comparison of the normal with the pathological. Both individuals give results so nearly identical that what is said of one is also true of the other. The range of hydrogen ion concentration is between 6.3 and 5.3, varying roughly with the protein intake, the average for W. W. P. being 5.7, for J. H. M. 5.8, values which are slightly higher than the one of 5.98 previously reported. As might be expected, the amounts of acid and ammonia increase considerably with the increase in protein. For this reason they can not fairly be compared to our earlier normal values, but otherwise call for little comment. Our chief interest lies in the behavior of the acid-ammonia ratio designated as R in our earlier papers. This ratio varies considerably in the two individuals but never exceeds 1.00, the average for the twelve days in W. W. P. being 0.785, with a range of 0.53 to 0.95, in J. H. M. 0.797, varying between 0.57 and 0.98, average values only slightly higher than the one of 0.75 found in normal individuals on a mixed diet.

A quite remarkably close agreement on same days is found in the ammonia-nitrogen ratio which varies between 7.5 on the low protein days to 4.0 when the protein intake is high. On one day only is there a marked discrepancy, in which W. W. P. has a ratio of 6.5 as against 4.8 in J. H. M. This close agreement between the two individuals is due unquestionably to the uniformity in amounts and articles of food.

III. RESULTS IN NEPHRITIS

In Table 2 we have assembled the cases of nephritis in order of the value of the ratio between acid and ammonia. The general average of all the observations of the individual cases is given in the table

because with the exception of Cases 4, 6, 20, 41, 43, 47 and 51, which have been added since, the detailed records of the acid factors appear in an earlier paper.[5] Identification of the cases in the earlier paper can be made by comparing the acid-ammonia ratio of the two tables.

In a few cases we were unable to obtain a full twenty-four hour amount of urine, but as the loss was relatively small, the ratio between acid and ammonia was determined. These cases together with other clinical data appear in Table 3.

Table 4 contains the general averages with the maximum and minimum variations as well as the normal average previously reported.

The total nitrogen in the urine was determined in Cases 4, 5, 6, 8, 18, 19, 20, 41, 43, 47 and 51, making it possible to compare the acid-ammonia ratio with the ammonia-nitrogen ratio in the urine. To this data we have added the noncoagulable nitrogen in the blood and collected the whole in Table 5.

Unfortunately only six of the cases studied in our series came to necropsy. The acid-ammonia ratio, clinical and anatomical diagnosis in these cases, are brought together in Table 6.

DISCUSSION

During the past few years there has appeared so much literature (much of which contains extensive reviews and bibliographies) dealing with "tests" of renal function, their value and significance, that any review of the subject here would be superfluous. A satisfactory survey of this general field is to be found in the publication of Blum[6] while the excretion of phenolsulphonephthalein, sodium chlorid, lactose, potassium iodid and water has been discussed by Fitz.[7] Since the appearance of Folin's method for the accurate and rapid determination of the noncoagulable nitrogen in small amounts of blood, several papers have been published on the value of increase in blood nitrogen as an aid in diagnosis and prognosis in renal disease. Recent publications with references to the work on this subject are those of Tileston and Comfort,[8] and Frothingham and Smillie.[9] Phenolsulphonephthalein

5. Henderson, L. J., and Palmer, W. W.: On the Several Factors of Acid Excretion in Nephritis, Jour. Biol. Chem., 1915, xxi, 37.

6. Blum, Victor: Nieren physiologie und functionelle Nierendiagnostik, Leipzig, 1913.

7. Fitz, R.: The Value of Tests for Renal Function in Early and Advanced Bright's Disease, Am. Jour. Med. Sc., 1914, cxlviii, 330.

8. Tileston, Wilder, and Comfort, C. W.: The Total Non-Protein Nitrogen and the Urea of the Blood in Health and in Disease, as Estimated by Folin's Methods, The Archives Int. Med., 1914, xiv, 620.

9. Frothingham, Jr., Channing, and Smillie, Wilson G.: The Relation Between the Phenolsulphonephthalein Excretion in the Urine and the Non-protein Nitrogen Content of the Blood in Human Cases, The Archives Int. Med., 1914, xiv, 541.

TABLE 2.—Data of Authors' Cases Assembled—

Case No.	Sex	Age	Clinical Diagnosis	Systolic Blood Pressure, mm. Hg	Urine			
					Phenolsulphonephthalein % in 2 Hrs.	Sp. Gr.	Albumin	Microscopical
1	M	36	Chronic nephrtis	190	Trace	1.012	T	No casts. Few W. B. C.
2	M	42	Chronic nephritis albuminuric retinitis.	180	Trace	1.017	S T	Occasional hyaline cast with fat adherent. R. B. C.
3	M	47	Chronic glomerulo	150	32	1.010	S T	Rare hyaline cast. Many R. B. C.
4	M	18	Chronic glomerulo; albuminuric retinitis.	180	.....	1.010	L T	Few granular casts, fat adherent. R. B. C.
5	M	22	Chronic glomerulo; albuminuric retinitis.	250	.....	1.015	T	Occasional granular cast. W. B. C.
6	M	33	Chronic glomerulo; albuminuric retinitis.	220	5	1.012	T	Many granular, a few fatty casts.
7	M	74	Arteriosclerosis (renal)...	205	15	1.018	T	Many hyaline and granular casts. W. B. C.
8	M	48	Acute nephritis	170	33	1.015	T	Granular and hyaline casts. Many R. B. C.
9	M	61	Chronic glomerulo; albuminuric retinitis.	230	.....	1.012	S T	Granular and fatty casts. Many W. B. C.
10	M	17	Chronic glomerulo	155	63	1.015	L T	Many granular and fatty casts. R. B. C. W. B. C. Few hyaline casts.
11	M	33	Acute nephritis	160	40	1.010	S T	Hyaline and granular casts. Many R. B. C.
12	M	45	Chronic glomerulo; albuminuric retinitis.	250	Trace	1.015	T	Hyaline and granular casts.
13	M	29	Chronic glomerulo; albuminuric retinitis.	200	11	1.010	S T	Rare granular casts.....
14	M	32	Nephritis (syphilitic); cirrhosis of the liver.	140	51	1.020	T	Many granular casts; few R. B. C.
15	M	29	Chronic glomerulo; albuminuric retinitis.	210	5	1.012	T	Many granular, few hyaline casts. Few R. B. C.
16	F	32	Chronic glomerulo	175	.....	1.016	L T	Hyaline and granular casts. R. B. C.; W. B. C.

—IN ORDER OF RATIO BETWEEN ACID AND AMMONIA

Urine							Remarks
		Acid Factors					
Number of Observations	Amount c.c. in 24 Hours	Hydrogen Ion Concentration	Acid N/10 c.c.	Ammonia N/10 c.c.	Acid + Ammonia	Acid ÷ Ammonia	
4	1,094	5.3	217	55	272	3.95	Scarlet fever followed by nephritis when young. Hypertrophied heart. Moderate edema. Chronic uremia. Died five months after leaving the hospital.
5	1,365	5.6	196	59	255	3.33	Scarlet fever when young. Nine weeks with edema. Two weeks uremic. Dyspnea. Died.
2	1,770	5.7	297	108	405	2.95	Frontal sinus at 35. For seven or eight years attacks of hematuria with mild uremia. Left hospital feeling better of one of these attacks.
7	2,280	5.5	350	118	468	2.97	For three weeks failing vision and puffy eyelids. Developed marked uremia, convulsions relieved by usual measures. Discharged from the hospital without symptoms save poor vision.
2	1,980	5.0	274	100	374	2.74	Etiology not clear. For six months increasing symptoms of uremia. Failing vision. At onset, edema. Died four days after observations.
4	900	5.0	194	75	269	2.60	Tonsillitis three years ago. Moderate alcohol. Mild uremic symptoms for six months. For two weeks failing vision, dyspnea and slight edema of eyelids. Cardiac hypertrophy, arteriosclerosis. Left hospital without symptoms.
2	1,346	5.0	265	128	393	2.07	A painter, lead colic twenty years ago. For a year increasing dyspnea and uremic symptoms. Cheyne-Stokes respiration. Cardiac hypertrophy, marked arteriosclerosis, edema of legs. Died two weeks after observations.
5	1,548	5.1	395	204	599	1.94	Alcoholic. Acute coryza for a week, associated with mild edema. Slight cardiac hypertrophy, palpable arteries. Improved.
4	1,131	5.9	144	75	219	1.93	Increasing uremia for six months. Dyspnea. Hypertrophy of heart, arteriosclerosis. Died one week after observations. Necropsy, arteriosclerosis.
9	1,189	4.9	316	168	484	1.89	Etiology obscure. Varying degrees of edema for six months. Slight uremia. Nonprotein nitrogen in blood 90 mg. per 100 c.c. Much improved. Died three months after leaving hospital.
5	2,571	5.1	307	165	472	1.86	Alcoholic. Hematuria and edema for two weeks. Improved.
5	912	5.1	193	107	300	1.80	Alcoholic. Scarlet fever and frequent sore throat. Dyspnea for a year. Uremia for five weeks. Left hospital against advice; moribund three days after observations. Hypertrophy of heart. Died shortly after leaving hospital.
18	2,640	5.2	472	275	747	1.72	Tonsillitis six years ago. One year mild uremic symptoms. Hypertrophied heart. No marked arteriosclerosis. Only slightly improved. Died one year later. Chronic interstitial nephritis.
19	868	5.2	318	195	513	1.63	Syphilis. Positive Wassermann. Three months with ascites and mild uremia. Slight hypertrophy of heart. Cirrhosis of liver. Slight improvement.
10	1,478	5.1	318	196	514	1.62	Nine years ago appendix abscess drained. Eight years ago albumin found in urine. No symptoms until three months ago, mild uremia. Some improvement. Died shortly after leaving the hospital.
4	1,930	5.4	217	137	354	1.58	Alcohol and lead (water). Uremia for a year. Convulsions preceding entrance to hospital in a comatose condition. Hypertrophy of heart. Mild arteriosclerosis. Improved. Died eight months later.

TABLE 2.—Data of Authors' Cases Assembled in—

Case No.	Sex	Age	Clinical Diagnosis	Systolic Blood Pressure, mm. Hg	Urine			
					Phenolsulphonephthalein % in 24 Hrs.	Sp. Gr.	Albumin	Microscopical
17	M	43	Chronic glomerulo	220	20	1.015	T	Rare hyaline cast.....
18	M	43	Chronic interstitial arteriosclerosis; albuminuric retinitis	245	5	1.010	T	Many hyaline and granular casts.
19	M	38	Chronic glomerulo	140	50	1.015	V S T	Many coarse granular casts, few hyaline casts. Rare R. B. C.
20	M	26	Chronic glomerulo; albuminuric retinitis.	200	32	1.012	S T	Few hyaline and granular casts.
21	M	61	Chronic interstitial	230	15	1.020	S T	Numerous granular casts. W. B. C.
22	M	57	Chronic nephritis	170	.....	1.013	T	Fatty and granular casts. R.B.C.; W.B.C.
23	M	15	Chronic glomerulo	160	70	1.024	V S T	Few hyaline and granular casts. Many R. B. C. Many W. B. C.
24	M	63	Cardiorenal disease	220	Trace	1.010	V S T	Many granular casts....
25	F	53	Cardiorenal disease; albuminuric retinitis.	200	.....	1.012	S P T	Nothing seen in sediment
26	M	36	Subacute glomerulo	150	.....	1.018	L T	Many epithelial casts. Few R. B. C.
27	M	29	Subacute glomerulo	150	.....	1.030	L T	Many fatty casts. W.B.C.
28	M	48	Chronic glomerulo; albuminuric retinitis.	235	48	1.015	0	Rare hyaline and granular casts.
29	M	19	Chronic glomerulo	170	.....	1.015	T	Many hyaline and granular casts. W. B. C.
30	M	30	Nephritis (arteriosclerotic?).	140	40	1.025	T	Occasional hyaline cast. W. B. C.
31	M	20	Chronic glomerula	130	30	1.030	T	Many fatty casts. R.B.C.; W. B. C.
32	M	15	Subacute Nephritis	110	55	1.030	L T	Many granular and fatty casts. W. B. C.
33	M	19	Subacute glomerulo	125	31	1.018	L T	Many granular casts. Many R. B. C. Some W. B. C.
34	M	41	Cardiorenal disease; chronic glomerulo; albuminuric retinitis.	170	Trace	1.015	S T	Few hyaline casts. W.B.C.
35	M	37	Chronic nephritis	130	40	1.013	V S T	Few hyaline and granular casts. W. B. C.

—Order of Ratio Between Acid and Ammonia—(*Continued*)

Urine							Remarks
		Acid Factors					
Number of Observations	Amount c.c. in 24 Hours	Hydrogen Ion Concentration	Acid N/10 c.c.	Ammonia N/10 c.c.	Acid + Ammonia	Acid + Ammonia	
3	1,005	5.9	282	193	475	1.46	Mild uremia for several months. Big heart and some arteriosclerosis. No improvement. Died four months after leaving hospital.
2	3,710	5.0	463	322	785	1.44	Moderate alcohol. Wassermann suspicious. For three months, increasing dyspnea and uremia. Hypertrophy of heart, arteriosclerosis. Slight edema. Left against advice; died one week later.
5	1,534	4.9	430	348	778	1.24	Alcoholic. Septic arm three years ago; no albumin in urine then; eight days acute coryza followed by edema. No uremia. Heart enlarged. Left hospital improved.
4	2,321	5.2	399	330	729	1.21	Etiology obscure; eight weeks of mild uremia which increased. Died three weeks after observations.
11	994	5.0	269	231	500	1.16	Marked arteriosclerosis. For six months dyspnea; slight edema. Heart not much enlarged. Slight improvement. Died shortly after leaving hospital.
4	935	5.0	231	213	444	1.09	Moderate alcohol. Brassworker; ten days' history of edema. Not uremic. Heart enlarged. Arteries palpable. Relieved.
2	640	5.1	224	205	429	1.09	Tonsillitis two months ago. Edema which has increased past two weeks. Improved. Died nine months after leaving hospital.
11	1,487	5.0	216	204	420	1.06	For six months increasing dyspnea, weakness with edema. Heart enlarged and dilated. Much relieved on digitalis. Fundi show arteriosclerosis. Died two months after leaving hospital.
6	1,398	5.8	294	278	572	1.06	For three years increasing dyspnea, dizziness, weakness. Heart not enlarged. Arteriosclerosis. Relieved.
3	1,035	5.1	472	450	922	1.05	Alcoholic. Edema and ascites for three weeks. Slightly enlarged heart. Much relieved.
7	440	5.7	222	226	448	0.98	Three months increasing edema with occasional uremic attack, convulsions. Heart not enlarged. Died with a terminal pneumonia one week after observations.
7	1,643	4.9	385	395	780	0.98	Alcoholic. Dyspnea on exertion, nocturia. Much enlarged heart. Arteriosclerosis. Some improvement before discharge.
5	992	5.2	371	384	755	0.97	History of nephritis four years ago. For three months mild uremia. Blurred vision but fundi normal. Heart not enlarged. Improved. A year later in good condition.
3	890	5.3	340	352	692	0.97	Three years ago acute nephritis; at the time mild uremia; well until one month ago; slight edema and mild uremia. Slight hypertrophy of heart. Improved.
2	405	5.4	264	270	534	0.97	Mastoid operation three years ago. Albumin in urine two years ago. Since then mild uremic symptoms. Negative physical.
5	335	5.4	188	198	386	0.95	Eight years ago for three months severe tonsillitis. Past four months increasing edema and pallor. Physical negative. Died some months later.
2	868	6.0	260	280	540	0.93	Etiology obscure. Purpura for ten days. No other symptoms. Improved.
28	953	5.2	207	224	431	0.92	Discharging ear for twenty-one years. Is a painter. Lead colic at 21. For eleven months increasing weakness, dyspnea and edema. Enlarged heart. Marked arteriosclerosis. Died two weeks after observations.
5	1,091	5.9	166	201	367	0.83	Nephritis (?) at 13. Headache for one year, much worse past three weeks. Heart not enlarged. Artery walls just palpable. Fundi normal.

TABLE 2.—Data of Authors' Cases Assembled in—

Case No.	Sex	Age	Clinical Diagnosis	Systolic Blood Pressure, mm. Hg	Urine			
					Phenolsulphonephthalein % in 24 Hrs.	Sp. Gr.	Albumin	Microscopical
36	M	41	Cardiorenal disease	210	28	1.020	V S T	Few hyaline and granular casts.
37	M	55	Chronic nephritis; arteriosclerosis.	230	20	1.016	V S T	Rare hyaline cast. W.B.C. R.B.C.
38	F	30	Chronic glomerulo	115	65	1.018	L T	Very few granular casts. Squamous cells.
39	M	28	Acute nephritis	140	30	1.025	L T	Many hyaline and granular casts. Many R. B. C. W. B. C.
40	F	45	Cardiorenal disease; albuminuric retinitis.	230	38	1.020	V S T	Rare granular and hyaline casts. W. B. C.
41	M	30	Chronic glomerulo; albuminuric retinitis.	200	49	1.015	T	Many hyaline and granular casts. W. B. C.
42	M	50	Sclerosis of kidney; arteriosclerosis.	240	10	1.014	S T	Few granular casts. W. B. C.
43	M	46	Chronic interstitial	260	20	1.015	V S T	Few hyaline and granular casts.
44	M	46	Arteriosclerosis; myocardial weakness.	150	41	1.012	V S T	Rare granular cast.....
45	M	19	Chronic nephritis	110	19	1.030	L T	Many hyaline, granular, fatty and epithelial casts. W. B. C.
46	F	43	Chronic nephritis; albuminuric retinitis.	210	33	1.022	L T	Many granular casts. W. B. C.
47	M	20	Subacute nephritis	160	70	1.010	S T	No casts were found.....
48	M	56	Chronic nephritis	210	.....	1.016	T	Few granular casts. W. B. C.
49	M	43	Chronic glomerulo	230	30	1.020	T	Numerous hyaline and granular, occasional epithelial casts. W. B. C.
50	M	25	Acute glomerulo	130	.....	1.025	L T	Many hyaline, granular and epithelial casts. R. B. C. W. B. C.
51	F	50	Chronic interstitial	260	42	1.020	V S T	No casts seen.....

—Order of Ratio Between Acid and Ammonia—(*Continued*)

Urine							
		Acid Factors					
Number of Observations	Amount c.c. in 24 Hours	Hydrogen Ion Concentration	Acid N/10 c.c.	Ammonia N/10 c.c.	Acid + Ammonia	Acid ÷ Ammonia	Remarks
9	1,300	4.9	408	497	905	0.82	Painter. Chronic lead poisoning. For two or three years, increasing dyspnea, precordial distress and edema. Enlarged heart. Arteriosclerosis. Little improvement. Died year later.
11	1,613	5.3	325	406	731	0.80	A year's dyspnea and edema which have been increasing. Very much enlarged heart. Marked arteriosclerosis. Fundi show arteriosclerosis. Some improvement. Died nine months later.
7	1,088	5.7	292	378	670	0.77	Albumin in urine since birth of child twelve years ago without symptoms. Came to hospital for arthritis. Heart and arteries not remarkable. A year later feeling well. No uremic symptoms.
3	916	5.3	455	596	1,051	0.76	Sore throat for three days with development of peritonsillar abscess. Acute nephritis with this. Heart and arteries negative. Improved.
5	1,358	5.0	193	256	449	0.75	For two months fatigue, paresthesias and slight right sided hemiplegia which had nearly all disappeared at time of entrance to hospital. Enlarged heart. Arteriosclerosis.
4	1,090	5.2	555	754	1,309	0.74	Etiology obscure. Two weeks swollen face and limbs. Wassermann suspicious. Heart slightly enlarged. Arteries not markedly sclerosed. In wards was mildly uremic. Slightly improved.
5	1,838	5.9	187	256	443	0.73	Six months' dyspnea, and paresthesias. Two slight cerebral hemorrhages. Big dilated heart. Arteriosclerosis. Some improvement. Died six weeks after leaving hospital.
7	1,936	5.5	320	442	762	0.72	Alcoholic. Three months of dyspnea and palpitation. Enlarged heart. Arteriosclerosis. Fundi show arteriosclerosis. Improved. Died two months later.
10	1,733	6.4	176	252	428	0.70	Dyspnea, weakness and puffy eyelids for two months. Enlarged heart and arteriosclerosis. Improved. Died a year later.
26	482	5.7	215	316	531	0.68	Always well. Five weeks increasing edema which was very marked at entrance. Physical except for edema not remarkable. Well year later.
6	796	6.0	175	261	436	0.67	For past six years two or three times a year has had brief attacks of dyspnea. Symptoms at entry were of cardiac decompensation and uremia. Heart enlarged, arteriosclerosis. Died two weeks after observation. Necropsy, arteriosclerosis.
6	3,433	6.4	268	397	665	0.64	No etiology. No symptoms. Albumin found in urine as result of examination for life insurance. Physical negative.
9	1,122	5.5	234	383	617	0.61	Three years' history of increasing dyspnea, headache and edema. Large heart, arteriosclerosis. Died in uremia and broken compensation two weeks after observations. Necropsy, arteriosclerosis.
3	1,090	4.9	358	586	944	0.61	Syphilis twelve years ago. Positive Wassermann now. For two months increasing but never severe uremia, edema. Heart enlarged. Arteries palpable but not tortuous. Improvement. Died two months later.
7	640	5.2	316	548	864	0.58	Two weeks mild uremia. Marked edema. Always well before. Physical shows nothing but edema. Marked improvement.
3	897	5.4	212	397	309	0.52	For two years, more or less, dyspnea. Recently mild uremic symptoms. Heart enlarged. Arteries sclerosed. Improved.

excretion, unquestionably the most convenient "test" of renal function, enjoys by far the most universal popularity and use; when it shows diminished output of color it apparently serves as an approximate estimate of kidney damage. There have been reported, however, several cases of severe nephritis in which the phenolsulphonephthalein excretion was *not* markedly reduced. On the other hand, recent writers seem to agree that any marked increase of the nonprotein nitrogen in the blood is of undoubted significance and probably involves fewer failures in detecting serious disease of the kidneys than

TABLE 3.—Cases in Which the Full–

Case No.	Sex	Age	Clinical Diagnosis	Systolic Blood Pressure, mm. Hg	Urine			
					Phenolsulphonephthalein % in 2 Hrs.	Sp. Gr.	Albumin	Microscopical
52	F	17	Chronic glomerulo	150	10	1.010	S T	Rare granular cast. R. B. C.; W. B. C.
53	M	23	Chronic glomerulo	240	.....	1.016	L T	Rare R. B. C. No casts
54	M	16	Chronic glomerulo	180	15	1.020	L T	Many granular casts. Many R. B. C.; W. B. C.
55	F	33	Chronic glomerulo; albuminuric retinitis.	210	.....	1.010	T	Very few casts. R. B. C.; W. B. C.
56	M	42	Chronic glomerulo	230	.....	1.008	S T	Few granular casts.....
57	M	29	Chronic glomerulo	130	40	1.020	S T	Hyaline and granular casts. W. B. C.
58	F	52	Cardiorenal disease; albuminuric retinitis.	210	0	1.010	S T	Rare granular cast. R. B. C.; W. B. C.

any other known test for renal sufficiency or insufficiency. Yet it may be pointed out here that there is frequently an increase in the nonprotein nitrogen in conditions other than nephritis. Satisfactory determinations of noncoagulable nitrogen requires not only much more than the average clinical laboratory equipment, but considerable skill in chemical technic. This fact makes the test less applicable to general clinical use. Still less information of diagnostic and prognostic importance seems to be available in the study of sodium chlorid, lactose, potassium iodid, and water excretion.

Given a disease which extensively damages the kidney, it is reasonable to expect that several of the many functions of the kidney may

be injured in varying degrees. As the final regulation of the alkalinity of the body, which we now know varies within extraordinarily narrow limits, falls on the kidney, and as a condition of acidosis has been shown to exist in the severe grades of renal disease, it is but natural and logical to investigate the various factors of acid excretion in this connection. The data in the tables represent such an investigation.

Among the factors of acid excretion the ammonia, in certain types of nephritis, appears to be most affected. In comparing the normal with the pathological data we have found it convenient to compute the

—Twenty-Four-Hour Urine Was Not Obtained

Urine			Remarks
Number of Observations	Hydrogen Ion Concentration	Acid + Ammonia	
9	5.3	3.00	Three months ago tonsillitis. One month ago noma. Past three weeks increasing weakness, uremia and slight edema. Heart not enlarged. Died one week after observations were made.
4	5.1	2.36	Etiology obscure. Well until two days before entering the hospital. Rapidly increasing uremia of sudden onset; coma and convulsions. Slightly enlarged heart. Fundi show edema. Died one week after observations. Necropsy, chronic glomerulo nephritis.
4	5.1	2.28	Etiology obscure. Three months' weakness and gradually increasing edema. While in wards developed pneumonia; had convulsions; died in uremia two weeks after observation. Fundi never showed changes.
5	5.2	2.14	Delivered of child three weeks ago after sudden onset of convulsions, nausea and vomiting. Increasing uremia since. Heart not much enlarged. Fundi show albuminuric retinitis. Died two weeks after observations. Necropsy, chronic glomerulonephritis.
3	5.1	1.26	Scarlet fever at 10. Peritonsillar abscess four years ago. For a year and a half tired and weak, for a year frequently. Losing weight. Eyesight failing. Enlarged heart. Palpable arteries. Not improved. Left against advice.
5	5.4	1.23	Pneumonia one year before entrance, albumin in urine ever since. For eight weeks edema of legs, headaches, poor appetite. Heart not enlarged. Fundi normal. Improved.
6	5.1	1.18	Loss of weight, color and strength for months. Mild uremic symptoms for eight weeks, has been increasing. Heart enlarged. No marked arteriosclerosis. Became more and more uremic. Left against advice unimproved.

ratio between acid and ammonia, hence the column in the tables, acid ÷ ammonia. The marked diminution in the relative as well as absolute amounts of ammonia in certain types of nephritis make it desirable to separate the cases into two groups, one with a high acid-ammonio ratio, the other with a lower or nearly normal ratio. In the former group we have included all cases above the ratio 1.40; the cases below this figure constitute the second group (Tables 2, 3 and 4). Although this separation is not quite a sharply defined one, there are relatively few intermediate cases. The number of observations on the respective cases is so variable that it has seemed best to record the means of the several observations on individual cases and to aver-

TABLE 4.—General Averages

		Blood Pressure	Phenolsulphonephthalein	Sp. Gr.	Number Cases	Amount of Urine	Hydrogen Ion Concentration	Acid	Ammonia	Acid + Ammonia	Acid ÷ Ammonia
I	Average of means	196	19	1.014	18	1,651	5.28	290	149	439	1.95
	Minimum mean[1]	140	Trace	1.010	..	900	5.90	144	55	219	1.44
	Maximum mean[1]	250	63	1.020	..	3,710	4.90	463	322	785	3.95
II	Average of means	173	33	1.019	33	1,194	5.40	280	350	630	0.80
	Minimum mean[1]	110	Trace	1.010	..	335	6.40	166	198	252	0.52
	Maximum mean[1]	260	70	1.030	..	2,321	4 90	472	754	1,309	1.24
	Normal average[2]	...	..	....	16	1.231	5.94	278	370	649	0.75

1. These values are selected from the general table so that acid + ammonia, and acid ÷ ammonia appear incorrect in the table. Both acid and ammonia values are individual low records from different cases.
2. Henderson, Lawrence J., and Palmer, Walter W.: The Several Factors of Acid Excretion, Jour. Biol. Chem., 1914, xvii, 305.

TABLE 5.—Total Nitrogen in Urine and Noncoagulable Nitrogen in Blood with Ratios

Case No.	Volume c.c.	Hydrogen Ion Concentration	Acid N/10 c.c.	Ammonia N/10 c.c.	Acid + Ammonia	Acid + Ammonia	Ammonia N	Total N	Ammonia Nitrogen Ratio	Blood Nitrogen Mg. per 100 c.c. Blood
4	1,840	5.7	294	80	374	3.68	0.112	8.8	1.3	132
	1,700	5.7	300	135	435	2.22	0.191	8.1	2.4	80
5	1,710	5.0	240	100	340	2.40	0.14	7.4	1.9	
	2,250	5.0	308	100	408	3.08	0.14	10.0	1.4	194
6	950	5.0	189	70	250	2.57	0.097	7.2	1.3	
	1,000	5.0	213	89	302	2.40	0.125	8.5	1.5	
	795	5.0	180	71	251	2.54	0.100	6.8	1.5	190
	855	5.0	203	70	273	2.90	0.097	7.1	1.4	
8	1,840	5.0	390	220	610	1.77	0.31	10.5	3.0	
	1,320	5.3	317	164	481	1.93	0.23	7.8	2.9	
	1,380	5.3	373	163	536	2.28	0.23	9.2	2.5	66
	1,660	4.9	480	202	682	2.37	0.28	11.9	2.4	
18	4,120	5.0	515	375	890	1.37	0.53	18.8	2.8	
	3,300	5.0	419	270	680	1.52	0.38	17.5	2.2	236
19	1,780	5.0	400	318	718	1.26	0.45	13.7	3.3	
	1,430	5.0	422	284	706	1.48	0.40	11.9	3.4	
	1,610	4.9	480	400	880	1.20	0.56	16.1	3.5	
	1,500	4.9	442	350	792	1.26	0.49	14.0	3.5	53
	1,350	4.9	405	345	750	1.17	0.48	14.8	3.3	
	1,280	4.9	378	327	705	1.15	0.46	13.5	3.4	
	1,375	4.9	438	405	843	1.08	0.57	15.1	3.8	
	1,670	4.7	510	605	1,115	0.84	0.85	17.6	4.8	
	1,810	4.7	620	715	1,335	0.87	1.00	19.5	5.3	62
	1,700	4.7	555	740	1,295	0.75	1.04	16.5	6.3	
	1,700	4.7	387	580	967	0.67	0.81	10.9	6.8	
	1,525	4.7	382	515	897	0.74	0.73	10.2	7.1	
20	2,265	5.1	400	310	710	1.29	0.43	9.9	4.3	76
	2,420	5.1	510	440	950	1.16	0.61	14.6	4.2	
	2,500	5.3	325	270	595	1.20	0.38	11.1	3.4	
	2,100	5.3	360	300	660	1.20	0.42	11.0	3.8	104
41	1,250	5.3	540	810	1,350	0.67	1.13	13.5	8.4	83
	1,320	5.2	640	684	1,324	0.94	0.96	11.8	8.1	
	1,040	5.1	535	520	1,055	1.03	0.73	11.8	6.2	
	1,840	5.0	510	1,000	1,510	0.51	1.40	10.5	13.0	116
43	2,075	5.2	405	470	875	0.86	0.66	12.2	5.4	
	1,900	5.6	304	570	874	0.53	0.80	12.7	6.3	88
	2,310	5.5	348	435	783	0.80	0.61	13.0	4.7	
	2,000	5.3	330	480	810	0.69	0.67	11.0	6.1	
	1,820	5.7	334	425	759	0.79	0.60	9.0	6.6	
	1,800	5.4	270	405	675	0.67	0.57	9.0	6.4	
47	2,600	6.0	230	400	630	0.58	0.56	11.6	4.8	
	3,700	6.7	220	320	540	0.69	0.45	6.2	7.3	
	4,100	7.0	82	400	482	0.17	0.56	6.5	8.6	
	2,800	6.9	140	250	390	0.56	0.35	4.1	8.5	53
	4,000	6.1	440	510	950	0.86	0.71	12.2	5.8	
	3,600	5.8	500	500	1,000	1.00	0.70	13.8	5.1	71
51	1,450	5.2	306	493	799	0.62	0.69	9.7	7.1	
	800	5.6	155	342	497	0.45	0.48	5.6	8.6	
	440	5.3	176	357	533	0.49	0.50	6.1	8.2	60

TABLE 6.—Data Concerning Necropsy Cases

Case No.	Acid ÷ Ammonia	Clinical Diagnosis	Anatomical Diagnosis	Description of Kidneys*
53	2.36	Chronic glomerulonephritis; chronic endocarditis of the mitral valve; hypertrophy and dilatation of heart.	Chronic glomerulonephritis; fibrous endocarditis of the mitral valve; hypertrophy and dilatation of heart; soft spleen; obsolete tuberculosis of the mesenteric lymph nodes.	Combined weight 236 gm. Capsule adherent in places. The surfaces generally finely granular and mottled with discrete and confluent pin-head sized yellowish to grayish areas. On section the cortical markings are obscured and the cortical substance shows discrete and confluent yellow to grayish ill-defined areas and mottlings. Over considerable areas the cortical substance is of a homogeneous opaque yellowish color. The cortex of each kidney is about 4 mm. wide. Microscopical: Considerable diffuse fibroid atrophy of the renal tissue. Atrophy of the glomeruli. Some glomeruli show proliferation of their endothelium as well as proliferation of the capsular epithelium. Arteriosclerosis is not marked.
55	2.11	Chronic glomerulonephritis; albuminuric retinitis; uremia.	Chronic glomerulonephritis; slight arteriosclerosis; hypertrophy and dilatation of heart; anasarca; edema of the limbs.	Weight of right, 156 gm. Capsule strips, leaving a smooth surface mottled with brownish red areas. The tissue is of good consistence; the pyramids are distinct from the cortex which measures 5 mm. The ends of the blood vessels are quite distinct. Weight of left kidney 92 gm. The capsule strips leaving a granular red surface. On section the tissue is rather tough, the markings are indistinct and the cortex measures 2-4 mm. The section surfaces show pale, dull, red pyramids and the cortices mottling with pale dull grayish and brownish areas. Microscopical: Marked degenerative changes in the tubules with considerable increase in the interstitial connective tissue. The glomeruli show well marked proliferation of the endothelium of their capillaries and occlusion of the latter by proliferated cells. Marked arteriosclerosis.
9	1.93	Chronic glomerulonephritis; hypertrophy and dilatation of heart; chronic passive congestion; mild uremia; acute pericarditis.	Arteriosclerotic nephritis; hypertrophy and dilatation of heart; chronic passive congestion; acute serofibrinous pericarditis; bronchopneumonia lower left lung; edema of the lungs; moderate arteriosclerosis.	Combined weight 254 gm. Capsules removable, leaving a very granular surface of a generally red color with fine gray mottling. Here and there a yellowish area about a pinhead in size is present. On section the cortices are 2-4 mm. inside. The cut surface is generally gray, red mottled and the markings obscure or absent. The renal arteries are large and the walls much thickened. Microscopical: Marked fibroid atrophic changes. The arteries, particularly the smaller ones, show marked sclerosis. There is some hyaline thrombosis. The glomeruli show some proliferation and fusion of their epithelium, but the appearances are not typical of chronic glomerulonephritis.

13	1.72	Chronic glomerulonephritis; albuminuric retinitis; uremia.	Chronic diffuse nephropathy; hypertrophy of left ventricle; edema of the lungs; edema of stomach and intestines; chronic hypertrophic gastritis; cerebral hemorrhage; fatty degeneration of heart.	Combined weight 190 gm. Both kidneys of the same size and general appearance. Each kidney contains a number of cysts varying in size from 3 to 0.5 cm. in diameter. The capsule is firmly attached but it strips off clean. The surface of the kidney is pale, roughly granular, due to small elevations averaging 2 mm. in diameter. On section the markings are obliterated. Both cortex and pyramids are atrophied, the average thickness of the cortex being 2 mm. The renal arteries show marked thickening and opacity of the intima. Microscopical: Sections of kidney show a high degree of arteriosclerosis affecting all the vessels, relatively the smallest vessels affected more than the large. The glomeruli show a high degree of atrophic and hyaline change; varying degrees of this hyaline change can be seen; in the slightest it shows as a thickening of the walls of the capillary loop; this is universally present. From this there is every change up to complete obstruction. There is very marked connective tissue increase with atrophy of the tubules. There seem to be comparatively few places in which all evidences of tubules have disappeared. There is great desquamation of the epithelium chiefly from the greatly dilated tubules in the cortex. It is difficult to account for this desquamation, because in many places the epithelial cells, though desquamated, show stainable nuclei. In the pelvis many of the collecting tubules are filled with masses of such desquamated cells. Hyaline casts are numerous.
46	0.67	Chronic nephritis; albuminuric retinitis; chronic uremia.	Arteriosclerotic nephritis; hypertrophy and dilatation of heart; arteriosclerosis; chronic passive congestion; thrombosis right iliac artery; infarct of spleen; acute and chronic colitis.	Right 70 gm. Left 135 gm. The capsule of the right kidney strips with difficulty. Here and there are rough puckered surfaces. Cortex measures 5-6 mm. On section the tissue is reddish brown and of slightly increased consistence. The markings are retained. The left kidney is similar to the right, only less scarred. Cortex 6-7 mm. Microscopical: Much arteriosclerosis. Areas of fibroid atrophy are present. The glomeruli are often enlarged and show doubtful proliferation of their endothelium.
49	0.61	Chronic nephritis; hypertrophy and dilatation of heart; anasarca; uremia; terminal infection.	Arteriosclerotic nephritis; hypertrophy and dilatation of heart; arteriosclerosis; chronic passive congestion; hydrothorax; anasarca and ascites; thrombi in right auricular appendage; chronic pleuritis.	Combined weight of the kidneys, 265 gm. The capsules strip leaving markedly granular, dark, gray, red surfaces which show a few small smooth walled cysts. Section shows increase in the connective tissue. The cortex and pyramids are made out, cortical markings obscured. The section surfaces show minute grayish areas and streaks; cut end of the vessels are prominent. Microscopical: Arteriosclerosis with atrophy and fibrosis of the renal tissue. The glomeruli do not show lesions characteristic of glomerulonephritis.

* The anatomical descriptions, with the exception of Case 13, are taken from the pathological records of the hospital, and have been verified in every instance by Dr. J. H. Wright. We are indebted to Dr. Henry A. Christian, of the Peter Bent Brigham Hospital, for the autopsy findings in Case 13.

age these means for the general averages rather than to obtain the average from the entire number of observations.

Group 1 includes the first eighteen cases of Table 2 and the first four of Table 3. Group 2 comprises all other cases. Because of incomplete amounts of urine in the subjects of Table 3 these values are not included in the general averages of the acid factors in Table 4. The acid-ammonia ratio in Group 1 varies between 1.44 and 3.95, averaging 1.95 as against a variation in Group 2 between 0.52 and 1.24 with an average of 0.80, which is very nearly the normal value. This very striking difference from the normal in Group 1 is not due to any increase in acid excretion, for the amount remains nearly normal, but to the diminution of ammonia excretion resulting finally in a reduction in the total acid. The hydrogen ion concentration in the two groups is 5.28 in the first and 5.40 in the second, a marked increase over the normal range of 5.94. The absence of an appreciable quantity of alkali in the form of ammonia in the ratio between acid and base in the urine helps to explain the high acidity in the first group. The cause of this increased acidity in the second group is not apparent, for the acid and ammonia values are quite normal. There is considerable difference between the two groups in the average volume of urine, 1,651 c.c. in the first and 1,194 c.c. in the second. The normal average found in our earlier investigations was 1,231 c.c. In both classes of cases, however, there is much variation. The factors which influence urinary volume are so many and so difficult to control that we do not care to attach much importance to this point.

Table 5 contains the detailed data of the several cases with urinary and blood nitrogen determinations. One might expect with the low amount of ammonia found in the high-ratio cases, a low ammonia-nitrogen ratio. This actually exists, as may be seen on examination of the data. Most references to metabolism in nephritis make little mention of urinary ammonia. In his book, "Metabolism and Practical Medicine," von Noorden,[10] briefly reviewing the subject, seems convinced that no difficulty exists in the excretion of ammonia salts, and calls attention to the fact that when any variation does occur it is in the nature of an increase rather than a decrease in their excretion. At one time Williams[11] and others considered the ammonia-nitrogen ratio of distinct value in differentiating between eclampsia and uremia, but Murlin and Bailey[12] have since pointed out its unreliability. Mar-

10. Von Noorden: Metabolism and Practical Medicine, English Translation, Chicago, 1907, ii, 434.

11. Williams: Pernicious Vomiting of Pregnancy, Bull. Johns Hopkins Hosp., 1906, xvii, 71. Bibliography.

12. Murlin and Bailey: Protein Metabolism in Late Pregnancy and the Puerperium, Jour. Am. Med. Assn., 1912, lix, 1522.

ischler[13] is the only investigator we have found who has made the observation that in chronic parenchymatous nephritis "in the stage of kidney insufficiency the excretion of ammonia is low, with improvement of the general condition of the disease, the daily amount of ammonia increases." In severe cases with a urinary nitrogen varying between 10 and 15 grams he found the ammonia-nitrogen ratio within the range of 0.5 and 2.8 per cent. The range of this ratio in normal metabolism as given by Folin[14] is 8.3 to 5.1 per cent. when the urine nitrogen is 14.8 to 18.2 grams, and 4.2 to 11.7 per cent. if the urine nitrogen is reduced to 4.8 to 8.0 grams.

In the first group this value is strikingly low, especially as the amounts of nitrogen in the urine are low, a condition accompanying the higher ammonia-nitrogen ratios in normal individuals. In Cases 4, 5 and 6 these ratios with a single exception are less than 2.0, although the urinary nitrogen does not exceed 10.0 grams and indeed is seldom more than 8.0 grams. With the diminution of the acid-ammonia ratio the ammonia-nitrogen figure increases until in Cases 43, 47 and 51 both ratios may be considered normal. Further evidence of marked disturbance in the kidney's function in eliminating the nitrogenous waste products is the high noncoagulable blood nitrogen in the high ratio cases. The acid-ammonia ratio and blood nitrogen could not be expected to vary exactly together, but in the tables there is a general tendency for the noncoagulable nitrogen to be lower as the acid-ammonia ratio approaches normal limits. Case 6 is interesting in that starting from a moderately high ratio, on a high protein diet, the urinary ammonia increased in relative amount. This case was one which was diagnosed clinically as acute nephritis, and might well represent one of the milder grades of renal damage. It is quite possible that the function of the kidney in respect to nitrogen elimination was improving during the course of the experiment or that the ammonia salts had reached such a high concentration in the blood that the kidney eliminated a sufficient amount to reduce the ratio between acid and ammonia. The latter explanation seems more probable in view of the fact that the blood nitrogen increased rather than decreased.

In the foregoing discussion of the nitrogen factors the abnormal variation has been attributed to the inability of the kidney to excrete the nitrogen constituents in a normal manner. We have not sufficient data at hand to make any other supposition. It is true, however, that in several cases not reported here we have found a high noncoagulable

13. Marischler, Julius: Ueber den Einfluss des Chlornatriums auf die Ausscheidung der Kranken Niere, Arch. f. Verdauungskr., 1901, vii, 332.

14. Folin, Otto: Approximately Complete Analyses of Thirty "Normal" Urines, Am. Jour. Physiol., 1905, xiii, 45.

nitrogen in the blood without any change in the acid-ammonia ratio. With this fact in mind one is tempted to speculate concerning the possibility of certain cases with high blood nitrogen in which renal insufficiency does not explain all the phenomena.

Certain relationships in the two groups between blood pressure, phenolsulphonephthalein excretion and specific gravity of the urine are not without interest. The blood pressure in the first group averages a little higher, 196 mm., than that of the second, 173 mm. The lower limit, 140 mm., was higher than in the second group, 110 mm., while the upper readings were about the same for both. The two hour phenolsulphonephthalein excretion was much lower in the first group, 19 per cent., than in the second, 33 per cent. The range in each was much the same, from a trace to 60 or 70 per cent. We would call attention to Case 10 in the first group which had a two hour phenolsulphonephthalein excretion of 63 per cent. and died in three months. Other things being equal, a lower specific gravity in the group with larger volume of urine would be expected. The average specific gravity in the first eighteen cases is 1.014, varying between the narrow limits of 1.010 and 1.020, while the range in the other cases was between 1.010 and 1.030, averaging 1.019. Albumin occurred in larger amounts in the high ratio cases. In Group 1, 5 had a large trace, 11 a trace, and 6 a slight trace of albumin, as compared with 9 a large trace, 7 a trace, 5 a slight trace, 9 a very slight trace and 1 in which it was absent in the second group.

We wish especially to call attention to the clinical diagnosis in reference to the variation of the acid-ammonia ratio. In Tables 2 and 3 with each case we have given the clinical diagnosis as it appears in the hospital records and in no case has any influence on the diagnosis been exerted by the authors of this paper. These diagnoses were made by one of the four visiting physicians to the hospital under whose charge the patient chanced to be.

The cases are collected in Table 7 with reference to the diagnoses and divided into groups as above, one with ratio greater than, a second less than, 1.40.

TABLE 7.—Diagnosis with Reference to the Acid-Ammonia Ratio

Group 1, acid-ammonia ratio greater than 1.40, includes Cases 1-18, 52-55.		Group 2, acid-ammonia ratio less than 1.40, includes Cases 19-55, 56-58.	
Chronic glomerulonephritis	15	Chronic glomerulonephritis	11
Chronic nephritis	2	Chronic nephritis	6
Acute nephritis	2	Cardiorenal disease	6
Arteriosclerotic nephritis	1	Chronic interstitial nephritis	3
Chronic interstitial nephritis	1	Arteriosclerotic nephritis	3
Syphilitic nephritis	1	Subacute glomerulonephritis	3
		Subacute nephritis	2
		Acute nephritis	2

In the group with high ratios there is a strikingly large proportion of cases supposed to be chronic glomerulonephritis, while many supposed to be some form of degenerative nephritis appear in the low-ratio group. Of the twenty-two high-ratio cases fifteen were definitely diagnosed as chronic glomerulonephritis. It is fair to add that the two cases in which the diagnosis of acute nephritis was made may have been merely acute exacerbations of a slowly progressing nephritis, while in the ones diagnosed as chronic nephritis there were some apparently borderline cases in which a definite clinical diagnosis was impossible. Syphilitic nephritis is obviously a diagnosis of convenience and may properly be considered under the head of chronic glomerulonephritis. All the cases in Group 1 were more or less uremic and must be deemed severe when one considers that eight of the twenty-two patients died in the hospital, 2 shortly and one each 3, 4, 5, 8 and 12 months, respectively, after leaving the hospital. In six cases our letter of inquiry was returned unclaimed.

In the second group the cases seem to be much more scattered among the different types of nephritis, but if the cases catalogued as "chronic nephritis," "cardiorenal disease," "chronic interstitial" and "arteriosclerotic nephritis" are taken together (as they probably deserve to be) exactly one half of the group is included. This grouping seems justifiable because the degenerative type of nephritis in most instances is indicated. Only a few of the cases in this group were diagnosed as chronic glomerulonephritis, and not one of the acute or subacute cases, either clinically or by the various functional tests employed, showed marked renal insufficiency. On the other hand, several among the cases of degenerative nephritis were uremic, and a few died in typical uremia. Our attempts to ascertain the present condition of the subjects in this group were less successful than in the preceding group. Thirteen letters were returned unclaimed. Five died in the hospital and one soon after leaving. One died in 6 weeks, 3 in two months, 2 in nine months and 3 a year after leaving. Eight of the cases are now known to be living and without symptoms.

Of the six cases coming to necropsy four had high and two low ratios (Table 5). The clinical diagnosis in all four of the high ratio cases was chronic glomerulonephritis, which in two was confirmed at autopsy. Of the other two, however, the anatomical diagnosis was, in one, arteriosclerotic and in the other chronic diffuse nephritis with a probable arteriosclerotic background. One of the cases, Case 9, was a man 61 years old whose kidneys showed on section considerable arteriosclerosis and some changes in the glomeruli which are considered in the autopsy report "not typical of chronic glomerulonephritis." The second high ratio case in which the clinical and anatomical diagnosis

disagreed was Case 13, a young man of 29 years. His kidneys on section revealed in addition to considerable arteriosclerosis, "glomeruli showing a high degree of atrophy and hyaline change." (See protocols.) The two low-ratio cases were each diagnosed clinically as chronic nephritis and at necropsy proved to have arteriosclerotic kidneys.

From the clinical evidence and limited pathological data at hand it seems safe to say that severe injury to the glomeruli is commonly accompanied by reduced ammonia excretion. In view of the difficulty in harmonizing clinical pictures and functional findings with anatomical diagnoses, we would refrain from giving the impression that in the values of the acid and ammonia we have a means of estimating the function of the kidney which conforms closely to the pathological changes. It is our purpose merely to point out an interesting phenomenon which accompanies severe grades of nephritis, usually of the chronic glomerulo type and which seems to us the more important because it is in these cases we invariably find a condition of acidosis.[5] It seems reasonable to suppose that the failure of ammonia to do its work in the regulation of the reaction of the body may be the cause of this condition.

CONCLUSIONS

We feel justified in drawing the conclusion that our cases of nephritis divide themselves into two groups possessing the following characteristics:

1. Cases in which the volume of urine is abnormally large, its acidity abnormally intense, and the total acid excretion much diminished (signs of a condition of acidosis which may be of renal origin). This diminished acid excretion is due exclusively to a never failing deficit in the urinary ammonia, for the value of A (acid) is, taking account of the intensity of acidity, precisely normal. In this group the late stages of glomerulonephritis predominate. It is usual to find high noncoagulable nitrogen, a much reduced phenolsulphonephthalein excretion, a high blood pressure, large amounts of albumin and low specific gravity of the urine.

2. Cases in which the mean urinary volume appears to be not far from normal, the acidity high and often very high, the total acid excretion often low, but not infrequently normal. The variation in this quantity is once more due to fluctuations in urinary ammonia. These cases suggest the idea that the cases of this group involve varying degrees of acidosis which are generally much milder than in the cases of Group 1. The degenerative type of nephritis and earlier stages of glomerulonephritis occur more frequently in this group. The noncoagulable nitrogen, though often increased, is not always strik-

ingly so. Phenolsulphonephthalein excretion is moderately reduced, or it may be markedly diminished; blood pressure varies widely, the specific gravity averages higher than in the previous group and albumin occurs in smaller amounts.

Group 1 appears to consist of an uncommonly sharply defined group of cases which, functionally at least, are of one type. Group 2 may well consist of either one or more classes of disturbance of renal function, including perhaps mild forms of the condition represented in Group 1.

STUDIES ON THE PATHOLOGICAL PHYSIOLOGY OF THE HEART

II. THE DYNAMICS OF AORTIC INSUFFICIENCY *

CARL J. WIGGERS, M.D.
NEW YORK

I. PREVIOUS EXPERIMENTAL WORK

Since Corrigan,[1] in 1832, first described and gave a logical explanation of the collapsing pulse often associated with aortic incompetency, clinicians and physiologists alike have given their interpretations of the circulatory changes inaugurated by a leakage of the aortic valves. In this, as in other provinces of medicine, it is important to differentiate between demonstrated facts and convenient assumptions to explain clinical observations. It is but natural, therefore, that the subject should have been the cause of considerable experimental investigation.

In 1878, Rosenbach[2] punctured the aortic valves in rabbits by a glass rod introduced through the right carotid. This caused no variation of mean arterial pressure. Cohnheim[3] confirmed this result but Goddard[4] obtained a fall of mean arterial pressure. De Jager,[5] in 1883, obtained no variation after puncturing the valve segments in dogs, but in rabbits he observed a fall. This he explained as due to the fact that a relatively greater insufficiency is produced in rabbits.

Kornfeld,[6] in 1896, extended the experimental work by recording in addition the pressure in the left auricle. In a considerable percentage of his experiments the mean arterial pressure fell. He considered three possible causes of this fall: a backward leak, a deficient cardiac contraction, and a reflex dilatation of the blood vessels. The rise of right auricular pressure in some experiments favored, he believed, the first possibility. The back-flow during diastole, he reasoned, elevated the intraventricular pressure and so raised the left auricular pressure. Those cases in which the auricular pressure was not elevated he

* Submitted for publication Feb. 19, 1915.

* From the Department of Physiology, Cornell University Medical College.

* The second of a series of experimental and clinical investigations of pathological cardiac conditions made by means of optically recording instruments.

1. Corrigan: Edinburgh Med. and Surg. Jour., 1832, xxxvii, 225.
2. Rosenbach: Arch. f. exper. Path. u. Pharmakol., 1878, ix, 1.
3. Cohnheim: Vorlesungen über allgemeine Path., ii, 38.
4. Goddard: Acad. Proefschrift, Leyden, 1879.
5. De Jager: Arch. f. d. ges. Physiol., 1883, xxxi, 215.
6. Kornfeld: Ztschr. f. klin. Med., 1896, xxix, 91, 344.

explained as due to a ventricular dilatation without increase in tension. Kornfeld also made the observation that the arterial pressure rapidly recovered from the fall and returned to its normal level, a fact conveniently attributed to a compensatory vasoconstriction.

It is evident that studies of the mean arterial pressure are not adequate to give any clear conception of the dynamics; in fact these experiments necessitate as many, if not more, theoretical assumptions than do the clinical signs and symptoms.

In 1908, Stewart,[7] for the first time, endeavored to study the dynamic changes during separate cardiac cycles. This was accomplished by recording simultaneously the volume curves of the ventricles by a cardiometer and tambour and the arterial pressure changes by a Hürthle membrane manometer, the maximal and minimal readings being controlled by maximal and minimal valved manometers.

On rupturing an aortic cusp by means of a valvulotome devised by MacCallum,[8] he found that diastolic pressure fell more than systolic, while, in some instances at least, the contour of the arterial pulse changed. The amplitude became greater and descending limb dropped more rapidly, corresponding to the typical collapsing pulse clinically observed.

The volume curve showed certain interesting but unexpected deviations—the rate of ventricular discharge estimated by the gradient of the downstroke was slower, the output per beat was increased only a trifle (5 c.c.) and the ventricular filling, occurring normally only in early diastole, continued throughout that phase. In spite of this extended period of filling, the ventricle was dilated less than is normally the case at the end of diastole, an observation that could be accounted for only by a greater tonicity.

Two facts are emphasized by this investigator, (1) that relatively little blood regurgitates during aortic insufficiency, and (2) that regurgitation can account neither for the large amplitude of the pulse nor for its collapsing character. The reason that little regurgitation occurs is logically explained as follows: The flow into the ventricle in diastole is determined by the size of the opening as well as by the pressure. At the end of systole, however, the pressure in the aorta is quite low (not more than 15 mm. above that at the end of diastole). Since the aortic leak is small as compared with the mitral orifice, blood flows more readily through the latter under low auricular pressure than through the narrow aortic leak under higher arterial pressure.

The larger amplitude of the arterial pulse and its collapsing character cannot be due to a regurgitation, according to Stewart, for, (1) the

7. Stewart: The Archives Int. Med., 1908, i, 102.
8. MacCallum: Bull. Johns Hopkins Hosp., 1906, xvii, 260.

great drop of pressure occurs before the dicrotic notch, and hence during ventricular systole, and (2) the collapsing pulse disappears if the peripheral resistance is increased. Stewart therefore interprets the collapsing pulse and great fall of diastolic pressure as due to a reflex dilatation of blood vessels, for (1) such changes are known to accompany vasodilatation and (2) irritation of the root of the aorta caused in his experiments a similar fall of diastolic pressure.

Experiments similar to those of Stewart were reported in 1909 by Zollinger,[9] who used rabbits, cats and dogs as experimental animals. He also found that the output per beat was practically unchanged after insufficiency, but that the diastolic distention was always increased. The systolic arterial pressure was slightly elevated or unchanged. The diastolic pressure was invariably reduced and the pulse pressure thereby increased. No typical change in contour could be recognized in the curves taken with a Hürthle torsion manometer.

In 1911, MacCallum[10] restudied the dynamics of aortic insufficiency by means of a perfusion system of such a nature that the heart intact within the thorax and inclosed within a cardiometer, ejected its fluid into a set of rubber tubes emptying into a reservoir from which, in turn, the right heart was fed. In this way the peripheral resistance was entirely eliminated.

The production of an aortic insufficiency by means of a valvulotome still caused a low diastolic pressure, while the systolic pressure remained unaltered. The amplitude of the volume curve indicating the systolic discharge increased and the ventricles dilated somewhat. Whereas, normally, the measured volume outflow from the rubber tubes and that calculated from the volume curve corresponded, after insufficiency, the output calculated from the volume curves increased while the measured flow remained unaltered. MacCallum concluded from this that the excess volume must have regurgitated back into the ventricle.

The greater systolic discharge, together with the lower tension of the arterial wall, account, according to this investigator, not only for the greater systolic filling and large pulse amplitude, but also for the low position of the dicrotic notch without the assumption of a peripheral dilatation.

II. CRITICAL ANALYSIS AND SIGNIFICANCE OF PREVIOUS WORK

It is necessary to consider critically to what extent it is demonstrated by previous experiments that the dynamic changes in aortic insufficiency are due either to aortic regurgitation or to peripheral vaso-

9. Zollinger: Arch. f. exper. Path. u. Pharmakol., 1909, lxi, 193.
10. MacCallum: Bull. Johns Hopkins Hosp., 1911, xxii, 197.

dilatation. It will no doubt be generally conceded that, if the production of a valvular lesion causes in an artificial circulation scheme in which peripheral changes are entirely eliminated changes similar to those in the body, it offers probable, though not absolute, evidence that the changes are not due to vascular dilatation. To be satisfactory as probable evidence, however, the records previous to the lesion should have a normal contour as well as a correct placement and reproduction of a dicrotic notch. After the lesion, the amplitude should be larger and the placement of the dicrotic notch should be lower. This is the case in the experiments reported by Marey on a mechanical model of the circulation, but not so in the records reported by MacCallum. If any conclusion could be drawn from a careful study of the *so-called dicrotic waves* before and after valvular lesions, as reported by the latter investigator, it would need to be that the dicrotic notch mounted higher on the descending limb during insufficiency (MacCallum, curves 2 and 5). As a matter of fact, however, owing to the use of inadequate apparatus, no oscillations resembling a dicrotic notch were recorded, but instead the inherent vibrations of the apparatus.

On the other hand, however, no satisfactory proof has been offered by Stewart that a dilatation occurs. The curves reproduced to show the possibility of producing a reflex vasodilatation on irritating the root of the aorta are clearly misinterpreted (Stewart, Figures 20 and 21). The fall of diastolic pressure was quite evidently due to a slowing of the heart[11] and not to a reflex dilatation. The proof rests entirely on the necessity of explaining (a) the great fall of arterial pressure previous to the dicrotic notch, and (b) the supposed return of the pulse contour to normal after aortic compression or the administration of epinephrin. To anticipate, it may be stated that the former can be explained on an entirely different dynamic basis, while the latter observation proves to be incorrect.

The evidence seems to be conflicting as to whether an actual regurgitation occurs. MacCallum found the amplitude of the volume curve increased. Stewart and Zöllinger obtained no change. Stewart found that at the end of diastole the ventricle was dilated less than normally; Zöllinger and MacCallum noted considerable distention.

It is questionable to my mind whether this mode of experimentation is reliable or conclusive. During the time interval required to produce the lesion the circulation may have been changed, and by the manipulation itself it is difficult not to disarrange the cardiometer enough to account for the changes. A more fundamental objection, however,

11. For a detailed discussion of the effect of the length of previous heart cycles on systolic and diastolic pressures of subsequent beats, see Wiggers, Jour. Exper. Med., 1914, xix, 12; also The Circulation in Health and Disease, Philadelphia, 1915, p. 71.

exists. The cardiometer is applied to two ventricles and records their volume changes simultaneously. Its use in the normal circulation is based on the presumption that the two ventricles functionate in a similar manner. Who shall dare to accurately analyze the composite curve obtained when one ventricle contracts as an after-loaded and the other as a loaded muscle? The results during insufficiency can at most show the changes in the volume output of the two ventricles. The fact that the amplitude fails to increase does not necessarily imply the failure of a regurgitation. It is conceivable, for instance, that a regurgitation may take place into the left ventricle, push the interventricular septum to the right and in so doing prevent the filling of the right ventricle by an amount equal to the increase regurgitation into the left.[12]

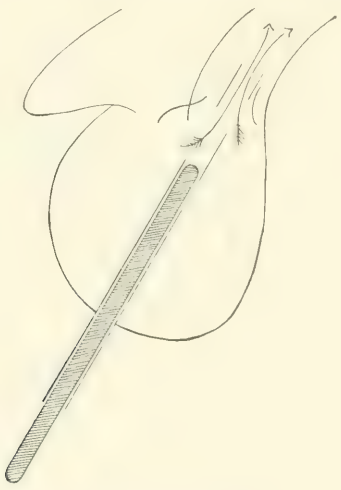


Fig. 1.—Diagram illustrating a simple method of producing aortic insufficiency in the expired heart.

A critical consideration, therefore, leads to the conclusion that clear and convincing proof has not yet been supplied by experimental work as to the dynamic changes brought about in insufficiency.

III. APPARATUS AND PROCEDURES

In this investigation the changes in the aortic and intraventricular pressures immediately after the production of valvular insufficiency were studied. The manometers, the recording apparatus and technic of their operation were essentially as described in a previous paper of this series.[13] Mean pressure was read in addition from time to time by

12. Cf. results of Henderson and Prince, Heart, 1914, v, 217.
13. The Archives Int. Med., 1915, xv, 77.

temporarily opening a side tube of the optical manometers connected with a damped mercury manometer.

The dogs were under chloretone anesthesia. The chest was opened after proper artificial respiration had been instituted. The pericardium was left intact as far as possible. Provisions for maintaining an effective auricular pressure, when desired, were at hand, as described in a previous paper.

Valvular insufficiency was produced in a temporary fashion on a principle similar to that previously reported in conjunction with Du Bois.[14] The apparatus and method are diagrammatically illustrated in Figure 1. A metal sound 10 cm. long and of 5 mm. bore, closed by a snugly fitting glass plunger is forced through the pericardium and ventricular musculature at the apex and entered into the mouth of the aorta so that the slot (2 cm. long, 2 mm. wide) on one end, palpated through the aorta, lies exactly at the level of the valves. When the plunger is withdrawn an insufficiency results. When it is pushed in, the valves close normally about the sound. The advantages of producing temporary valvular lesions in this manner over tearing or cutting the valves are: (1) the lesion can be produced while tracings are taken and the apparatus remains undisturbed; (2) the degree of insufficiency can be controlled and gaged by the size of the opening; (3) normal controls as to whether the circulation has changed for other causes can be obtained after the lesions have been studied; (4) the method is simpler and more certain, requiring no preliminary practice. It is used by the students in the laboratory course of clinical physiology under Dr. Du Bois, who produce one lesion after the other on the same animal.

The order of experimentation has, except for special reasons, been as follows:

1. Record of subclavian pressure curve by a calibrated optically recording manometer.
2. A second record after opening the chest.
3. A normal record of the subclavian pressure alone or in combination with left intraventricular pressure after the circulatory conditions which it is desired to study have been produced.
4. A record of same after inserting the sound into the aortic orifice.
5. Records during temporary insufficiency.
6. Records after normal valve action has been restored.

IV. THE EFFECT OF AORTIC INSUFFICIENCY ON THE CONTOUR OF THE ARTERIAL PRESSURE CURVE

Eighteen experiments have been performed. A few segments of records are analyzed as representative of the dynamic changes recorded.

14. Wiggers and Du Bois: Proc. Soc. Exper. Biol. and Med., 1913, x, 87.

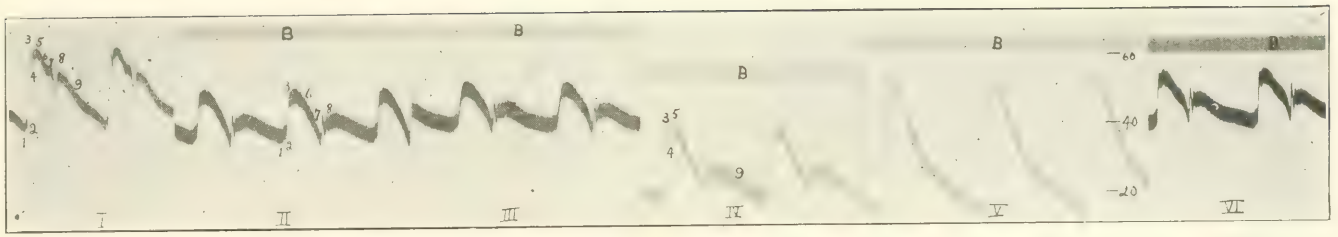


Fig. 2.—Six segments of records of subclavian pressure. *I*, normal, chest closed; *II*, normal, chest opened; III, sound introduced, mild stenosis; *II'*, mild degree insufficiency; *I'*, marked insufficiency; *I'I*, normal after; *B*, base line (description in text), which remains unchanged.

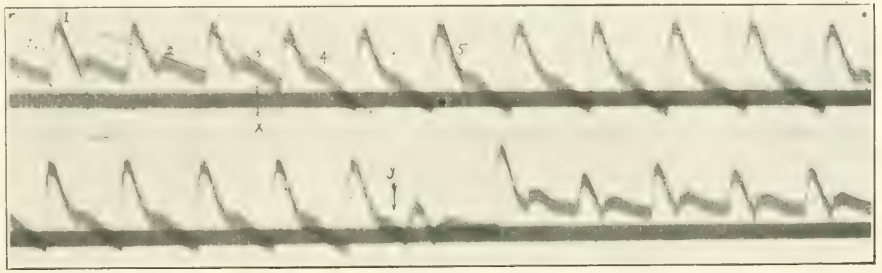


Fig. 3.—Continuous record of subclavian pressure during production of insufficiency at *x* and restoration of normal conditions at *y*. First two beats after *y* are due to accidental extra systole and postcompensatory pulse beat after. The third wave after *y* shows normal again.

a. Arterial Curves Obtained in Cases with Impaired Systolic Output

Experiment C 59, Feb. 16, 1914 (Figure 2).

I. Subclavian pressure recorded by optical manometer (N = 118); mean pressure 74 mm. mercury. The curves show all the details described by Frank, namely, 1-2, preliminary oscillations during the isometric period of systole; 2, 3, 4, preliminary oscillation of the arterial blood column; 5, systolic summit; 6, systolic decline; 7, end of systole and beginning of incisura; 8, vibration of closing valves (amplitude = 15 mm.); 9, gradual decline during diastole.

II. Same, immediately after opening the thorax; estimated hemorrhage, 10 c.c. The systolic pressure has fallen more than the diastolic; the primary wave (2, 3, 4) has almost disappeared; the systolic fall (6) is more marked and the pressure at the end of systole is low. The incisura (7) is more gradual in its drop and the valve vibrations are slightly increased in amplitude (16 mm.) but the period is unchanged.

III. Same, after inserting sound. A slight stenosis has been produced in this case, as is shown (a) by the more gradual rise of the ascending limb (causing the broader band of light); (b) by the round top and entire absence of any trace of the primary oscillation.

IV. Mild aortic insufficiency combined with previous slight stenosis. Heart rate exactly the same. The changes observed are: The primary oscillation (1-2) is entirely absent; the primary oscillation (3-4) returns; the systolic summit is very slightly higher (compared to base line, B); the pressure at the end of systole (7) is slightly lower; the rate of diastolic drop (9) is more rapid and the pressure at the end of diastole is much lower.

V. Marked insufficiency. Same general changes as in IV but more pronounced. Systolic pressure is much higher and diastolic pressure much lower (compared to base line, B). The primary oscillations (2-3, 4) are larger.

VI. Normal curve taken immediately after sound was withdrawn from the valve opening. In comparing this curve with II, it is evident that the condition of the heart and circulation has improved rather than suffered as a result of manipulation. Both systolic and diastolic pressures are higher than in II, while the curve approximates more nearly that obtained from the unopened chest.

The experiment was repeated in still another way. While the record was being taken, as shown in Figure 3, an insufficiency was suddenly produced. This occurred early in diastole, as shown by the arrow. Immediately, the slope of the diastolic portion became steeper. In the first beat after insufficiency the systolic summit is slightly lower; in the second beat the primary oscillation is indicated, the upstroke becomes steeper (narrower line) and the systolic summit mounts to its normal level.

Comments: It is apparent that the mere act of opening the thorax at once alters the pressure relations in the aorta. The fact that the systolic pressure decreases more than the diastolic, as shown in the second segment of Figure 2, indicates that the change is due to a diminished output in consequence, probably, of the decreased effective pressure in the left auricle. *It may be pointed out that the effects of valve lesions have probably been studied only under these abnormal conditions by those investigators who applied cardiometers to the heart.* Such experiments, though carried on during a hypodynamic state are not without interest, however, as they are presumably typical of a certain class of clinical cases in which an insufficiency associated with a mild stenosis supervenes when the effective venous pressure is unusually low. Paradoxical as the statement may sound, it is evident that from a dynamic point of view an insufficiency is

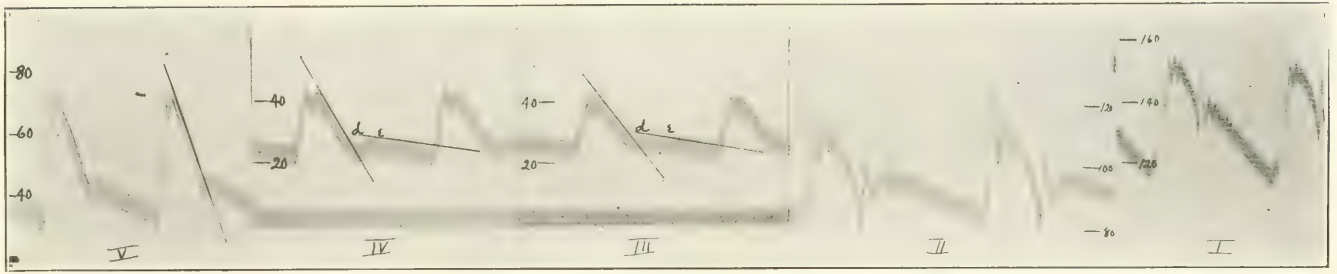


Fig. 4.—Five segments of records of subclavian pressure. *I*, normal, chest closed; *II*, normal, chest open; *III*, same during poor action of heart; *IV*, marked insufficiency; *V*, insufficiency during epinephrin.

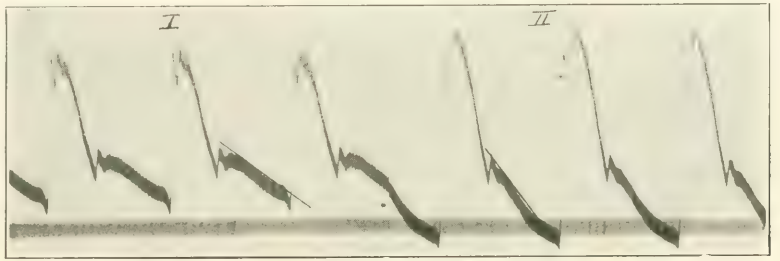


Fig. 5.—Two segments of records of subclavian pressure showing effect of aortic insufficiency when peripheral vessels were previously dilated.

apparently beneficial in such instances. The curves of the central arteries, which were rounded and devoid of secondary oscillations, such as are indicative of an efficient circulation, regain many of their normal features, such as the primary oscillation, the larger amplitude and sharp incisura (IV and I in Fig. 2). As far as the slope of the descending limb is concerned, the pressure falls more rapidly during systole, making the pressure reached at the end of systole somewhat lower, but the greatest fall occurs during the diastolic portion, and this accounts for the very low pressure at the end of diastole.

b. The Arterial Curves Obtained When the Muscular Power Is Impaired

Experiment C 62, February, 1914. Figure 4 (curves mounted in order from right to left). Subclavian pressure curve with very sensitive manometer (N = 50). The lower frequency probably accounts for the large primary oscillation and valve vibrations.

I. Chest closed, mean pressure 144 mm. Curve shows all the normal details and additional vibrations of the sensitive undamped instrument.

II. Chest open — mean pressure 78 mm.

III. After marked artificial respiration (acapnia ?) when the heart had become weaker and the output small — mean pressure 25 mm. The arterial curves lose all their normal characteristics.

IV. Aortic insufficiency of marked degree. The amplitude of the pressure change becomes greater, the systolic pressure is higher and evidence of a primary oscillation reappears. The diastolic pressure is slightly lower, the gradient of the slope is more rapid both in systole and diastole. The normal curve taken after this resembles the curve shown in III.

V. Epinephrin during insufficiency. Both systolic and diastolic pressures rise, but the pressure curve is still of the collapsing type. Mean pressure 50 mm. The primary oscillation becomes more prominent, giving the top a bifurcated appearance (pulsus bisferiens). *During systole the pressure falls more steeply in spite of intense vasoconstriction.* This leads one to infer that the rate of systolic fall is not governed by peripheral constriction or relaxation, but by the height of pressure at the beginning of systole.

Comments: When, in addition to a low venous pressure, the inherent response of the ventricular muscle is poor, aortic insufficiency does not cause variations of great amplitude. Both systolic and diastolic pressures are low. Neither a distinct incisura nor valvular after-vibrations occur. The systolic summit shows a distinct notch. As far as the changes in the descending limb are concerned the fall during systole is more rapid, but the diastolic slope remains unaltered. This is the case whenever the pressure during entire diastole is very low. Epinephrin intensifies the collapsing nature of the pulse.

c. The Arterial Curves Obtained When the Venous Supply and Functional Power Are Normal but the Peripheral Resistance Is Low

Experiment C 60, Feb. 18, 1914. Figure 5. Dog under chloretone anesthesia, slow saline infusion.

I. The curve is taken with the chest open. Venous pressure equals 50 mm. in the left auricle. Nitroglycerin previously administered. The curve shows all the details of a normal pulse except that the incisura is not so sharp, the amplitude is large and the pressure falls rapidly during systole.

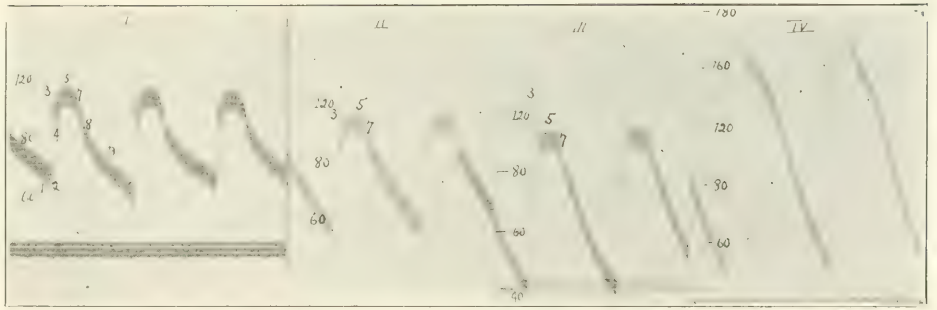


Fig. 6.—Four segments of records showing the effect in *II* and *III* of insufficiency when output and resistance are normal. *IV*, same after adrenalin. Compare all pressures to base line at bottom.

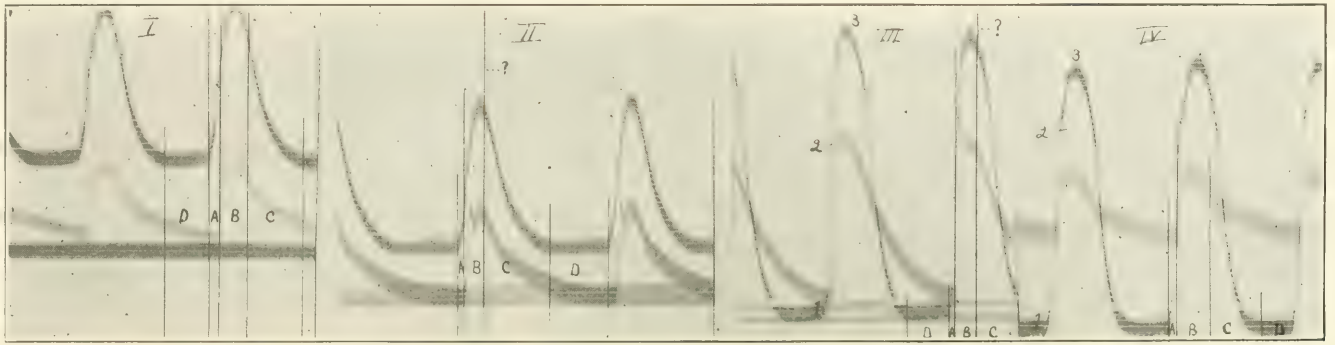


Fig. 7.—Four records of left intraventricular and subclavian pressures. *I* and *IV*, valves intact; *II* and *III*, aortic insufficiency.

II. Curve during insufficiency. All the details of the low resistance pulse are retained with the exception of the preliminary oscillations (1, 2, 3) occurring during the isometric period. The oscillations are all increased in amplitude, the systolic summit rises higher and the diastolic pressure reaches a lower level than the normal.

Comments: When the peripheral vessels are dilated and the output of the heart is normal, records of the largest amplitude are obtained, for, added to the tendency of a low resistance to increase the amplitude, is the effect of the lesion. The amplitude becomes greater because the systolic pressure is elevated and the diastolic pressure lowered. While the gradient of the pressure fall increases somewhat during systole, the most rapid drop responsible for the low pressure at the end of diastole occurs after the incisura.

d. Cases with Normal Output and High Peripheral Resistance

Experiment 6. Feb. 24, 1914. Figure 6. After opening the thorax the artificial respiration was reduced to a minimum, the dead space increased and a slow saline infusion continued. The thoracic aorta was slightly compressed by a special clamp.

I. Mean pressure, 106 mm. mercury. Curve shows characteristics very similar to those of the curves from the unopened thorax. The broad systolic summit is accounted for by the higher resistance. The numbers correspond to those of Figure 2. The valvular vibrations are reduced owing to a higher pressure.

II, III. Two degrees of insufficiency. The preliminary oscillations 1, 2, disappear, the primary vibrations (3) are larger and the top is sustained during systole (5), as in normal curves. The rapid drop at the incisura occurs in a normal manner. The change occurs in diastole beginning at 7. The fall is very rapid and complete so that the diastolic pressure is lower. The systolic summit also falls but not in proportion.

IV. After administration of epinephrin, 1 c.c. of a 1:50,000 solution. The entire curve has a larger amplitude. The ejection is more rapid but the contour of the systolic portion remains unaltered. The diastolic drop is more rapid, on the other hand, and of greater magnitude, indicating a greater regurgitation.

Comments: This type of experiment is comparable to clinical cases of insufficiency in which good heart action is associated with some peripheral sclerosis increasing the total arterial resistance. In such cases the change in contour is almost entirely limited to the diastolic portion. Increasing the activity of the heart, as by the injection of epinephrin, does not modify the contour of the systolic portion but increases the amplitude of the entire curve.

e. The Effect of Aortic Insufficiency on the Contour of the Intraventricular Pressure Curves

Experiment 62. Feb. 25, 1914. Figure 7. Simultaneous tracings of subclavian and left intraventricular pressures. The relative position of points is shown at the start of the second and third records.

I. Normal peripheral resistance, low venous pressure and a systolic output less than normal. Mean pressure, 40 mm. The intraventricular pressure curve possesses the smooth character common to curves in which the initial intra-

ventricular tension is low (see previous paper). The isometric rise (A) merges almost imperceptibly into the ejection period (B). C represents the active relaxation, and D the interval of diastasis.

II. Aortic insufficiency with changes typical of the arterial pulse in this condition — mean pressure 24 mm. The cycles are equal to those of I. An isometric period (A) persists but is shortened. The end of the ejection period is difficult to determine. Important, however, is the fact that the ejection period plus the relaxation period (B + C) remains unaltered, while the period of diastasis (D) is longer.

III. Aortic insufficiency after injection of saline and 2 c.c. of a 1:100,000 solution of epinephrin. The intraventricular record was shifted in relation to the base line in order to record the full amplitude. The most important change is that the intraventricular curve no longer remains smooth. A distinct bend in the ascending limb (2) separates the isometric and ejection periods. The systolic summit (3) rises exceedingly high.

IV. Normal curves during the action of epinephrin. The initial tension (1) is reduced but not in corresponding measure as the subclavian pressure is raised. The period of the isometric rise (A) is longer and terminates at a higher level (2). The summit (3) is lower, the period of active relaxation (C) is unchanged, but diastasis (D) is shortened in cases in which the same heart cycle is retained.

Comments: In regard to the intraventricular pressure curves, as compared with the arterial curves, aortic insufficiency causes, not an abolition, but merely a shortening of the isometric phase. The maximum pressure, as in normal hearts, is determined largely by the initial pressure which, during insufficiency, may be greater. This constitutes the immediate reserve mechanism of the heart which causes directly after the production of a lesion, a more vigorous output. In consequence of this continued greater activity, the ventricle probably hypertrophies. According to these conceptions, hypertrophy is a sequel, not a cause of an unusually large output of the heart in aortic insufficiency. The effect of insufficiency on the duration of diastasis, as revealed in these records, is precisely the opposite of the results of Stewart. Whereas this investigator observed a shortening or abolition of the period of diastasis, these curves show a distinct lengthening. Neither does it appear that the rate of active relaxation has altered. The evidence that the tonus changes was entirely lacking in all experiments.

V. SUMMARY AND DISCUSSION

The tracings obtained in this investigation, of which the illustrations are but selected segments, indicate that the details of the curves in aortic insufficiency depend, to a considerable degree, on the condition of the heart and the peripheral vessels at the time that the lesions are produced. Since all of these combinations and others are probably found in clinical cases, it may be well to summarize the chief changes in tabular form.

TABLE OF CHANGES IN AORTIC INSUFFICIENCY

Conditions of Circulation	Systolic Summit	Systolic Portion of Fall (to Onset of Incisura)	Diastolic Portion of Fall (After Valve Vibration)	Lowest Diastolic Pressure
Low venous pressure, decreased systolic output. Inherent action of heart good.	Higher; more peaked.	Gradient, somewhat steeper.	Gradient, markedly steeper.	Greatly depressed.
Inherent power of heart poor.	Slightly higher; bifurcated peak.	Gradient, steeper.	Rate of fall unaltered.	Unaltered.
Systolic output normal; peripheral vessels dilated.	Higher; increased amplitude of primary notch.	Unaltered.	Gradient, steeper.	Greatly depressed.
Systolic output normal. Peripheral resistance somewhat increased.	Slightly decreased.	Unaltered.	Gradient, much steeper.	Greatly depressed.

While the details of the curves vary under different conditions of the circulation, they show, in addition, certain constant changes, the significance of which can be definitely stated in dynamic terms. These changes it is desirable to discuss more at length.

1. During diastole, the pressure invariably falls more rapidly.[15] Indeed, this is the chief fall accounting for the low diastolic pressure. The contrary records obtained by a Hürthle membrane manometer from animals and by clinical sphygmographs from man, must be attributed to faulty apparatus which exaggerates the systolic portion by fling and cannot record the events which distinguish diastole from systole.

Since the change in diastolic slope occurs within the interval of a single heart cycle and is independent of dilatation of vessels by nitroglycerin and constriction by epinephrin, it is evidently associated with the effect of the lesion itself. It is, however, quite unnecessary to assume that a large quantity of fluid regurgitates. It is essentially the *pressure back-flow* that it is important to recognize.

Quite contrary to expectation, the intraventricular pressure curve during diastole shows no deviation in its contour. The curve merely

15. The only exception occurs when the diastolic pressure is very low, owing to a very small output.

fails to return to its normal level and in diastasis undergoes no further elevation, though arterial pressure continues to fall. The regurgitation of pressure occurs early after relaxation. The initial tension, that is, the tension to which the ventricle is submitted at the onset of the next ventricular contraction, is therefore never elevated so as to even approximate the intra-arterial diastolic. No detailed explanation can be offered without further investigation.

2. The preliminary oscillations normally present during the isometric period fail to occur. This is most readily explained by assuming that the ejection begins immediately after the onset of ventricular contraction. This would accord with the *current view* of the dynamics of this lesion. It is commonly recognized that the normal heart contracts as an *after-loaded muscle,* i. e., as a muscle which raises a weight so supported that it exerts its force only during the period of action. In other terms, the ventricle normally requires an isometric interval during which the pressure is raised sufficiently to open the semilunar valves and cause its ejection of blood. During aortic insufficiency, on the other hand, it is generally supposed that the ventricle is exposed to the full load of the aortic pressure during diastole and therefore contracts as a muscle from which a weight is permanently suspended. It has no isometric period, but the blood is ejected at once.

A careful comparison of the aortic and intraventricular records indicates that this view is not precisely correct. The curves shown in Figure 7, for example, clearly indicate that an isometric interval exists but that it is shortened. This is accounted for by the observation already pointed out, namely, that although the initial intraventricular tension at the onset of systole is somewhat greater during insufficiency, it is always less than the diastolic pressure within the aorta. Hence a time interval is required to elevate the intraventricular pressure to the level of the aortic diastolic pressure. The relatively low diastolic pressure and the more rapid elevation of intraventricular pressure combine to make the isometric period short. The failure of preliminary oscillations may be due to the fact that the small rise of pressure which is necessary before ejection occurs is not sufficient to cause a bulging of the valve segments.

3. The pressure rises more rapidly and the primary peak is augmented or is reestablished when absent previous to the production of the lesion. In order to understand the more rapid rise of the curve and the augmentation of the primary oscillation, it is necessary to bear in mind what may be termed *the dynamic law of the ventricle.* It has been shown by Frank in the case of the frog's ventricle, by me for the right ventricle of mammals and by Straub, Starling and Piper for

the left ventricle, that the rapidity of the tension rise, the maximum height which the intraventricular pressure reaches, as well as the vigor of ejection, are determined, within limits, by the initial tension within the ventricle. It has already been pointed out that without producing any deviation in the pressure curve, the initial tension is greater in aortic insufficiency. The ejection period also begins at a lower level, owing to the lower diastolic pressure. Consequently, a larger quantity of blood is ejected into the aorta and with greater rapidity than is normally the case. It should be recalled that the primary oscillations are due (Frank) to the vibration that the entire blood column undergoes when suddenly ejected. If, for any reason, the vigor of ventricular ejection becomes small, the primary oscillation may entirely disappear.

The production of an aortic insufficiency by increasing the vigor of discharge at once acts to restore or augment the primary oscillation. The ejection may be so sharp that several vibrations occur, as in Figure 6, III and IV.

4. The more vigorous ejection of a larger quantity of blood in the earlier part of systole also raises the systolic summit to a higher level and accounts for the high systolic pressure. It is apparently not necessary to assume the existence of a cardiac hypertrophy or an arteriosclerosis in patients in order to explain this high pressure.

5. The high systolic summit once reached is not maintained, but drops away very rapidly during systole, so that, at the beginning of the incisura, the pressure is often lower than the normal. That this is not due to an increased peripheral flow brought about by vasodilatation is evident from the facts (1) that the change occurs too rapidly (within a single beat), (2) that it is present when the vessels are previously dilated by nitroglycerin and (3) that it is intensified rather than prevented by epinephrin. It is probably explainable by the fact that, while a larger quantity of blood is ejected with greater vigor early in systole, the total systolic discharge is not much increased. Consequently, during the latter portion of systole, less blood per unit time is actually ejected, and the peripheral flow exceeds the cardiac output per unit time. It is therefore more pronounced when the vessels are dilated and entirely absent when the aorta is clamped and the area of peripheral flow restricted.

CONCLUSIONS

From these considerations it may be concluded (1) that the characteristic changes of the pressure curve in the central vessels, as recorded by optical manometers, cannot be explained by any reflex vasometer mechanism set in operation by the production of an insufficiency, and (2) that the dynamic changes are accounted for by the

fact that an aortic regurgitation increases the initial intraventricular tension, owing to a regurgitation of pressure during diastole; this, in turn, causes a more vigorous ejection of a larger blood volume in the early portion of the next systole. This may be accompanied by an actual decreased ejection during the latter portion of systole, thus, at once, accounting for the facts (a) that the systolic decline becomes steeper and (b) that the total systole output may not increase appreciably beyond normal.

First Avenue and Twenty-Eighth Street.

CONTENTS OF VOLUME XV

APRIL, 1915. NUMBER 4

MAY, 1915. NUMBER 5. PART 1

MAY, 1915. NUMBER 5. PART 2

JUNE, 1915. NUMBER 6

CONTENTS

THE ARCHIVES OF INTERNAL MEDICINE is issued monthly. Two volumes are published annually.

Manuscripts offered for publication and other correspondence relating to the editorial management of THE ARCHIVES should be sent to Dr. Joseph L. Miller, 122 South Michigan Boulevard, Chicago. Communications regarding subscriptions, reprints, etc., should be addressed to THE ARCHIVES OF INTERNAL MEDICINE, American Medical Association, 535 North Dearborn Street, Chicago.

Footnotes and bibliographies should conform to the style of the Index Medicus or to that of the Index-Catalogue of the Surgeon-General's Library. It is specially requested that the titles of articles and the exact date of issues, year, volume and page, be given in each case. Contributors are requested not to furnish bibliographies which are unnecessary, or mere clerical compilations. On receipt of 10 cents the American Medical Association will send a pamphlet entitled "Suggestions to Medical Authors," containing suggestions to authors on the preparation of manuscripts, rules for footnotes and bibliographies and a list of several hundred medical periodicals of the world with approved abbreviations of each.

Authors will receive one hundred reprints free; additional copies may be obtained at cost.

The domestic subscription price per year of two volumes is $4.00; Canadian rate $4.25; foreign rate $4.40 (18 shillings, 18 marks, 22 francs); including postage. Single copies 50c postpaid.

Make checks, etc., payable to the AMERICAN MEDICAL ASSOCIATION.